PCT

WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 :
C07C 323/22, 323/60, 323/62, A61K 31/16, 31/255

A1

(11) International Publication Number: WO 97/42168

(43) International Publication Date: 13 November 1997 (13.11.97)

(21) International Application Number: PCT/GB97/01164

(22) International Filing Date: 29 April 1997 (29.04.97)

(30) Priority Data:
96400958.3 6 May 1996 (06.05.96) EP
(34) Countries for which the regional or international application was filed: FR et al.
96402032.5 25 September 1996 (25.09.96) EP
(34) Countries for which the regional or international application was filed: FR et al.

(71) Applicants (for all designated States except US): ZENECA LIMITED [GB/GB]; 15 Stanhope Gate, London W1Y 6LN (GB). ZENECA PHARMA [FR/FR]; Centre de Recherches, Z.I. La Pompelle, Chemin de Vrilly, Boîte postale 1050, F-51689 Reims Cedex 2 (FR).

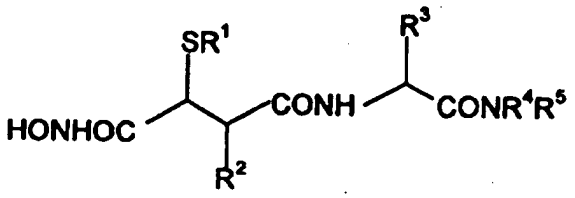
(72) Inventors; and
(75) Inventors/Applicants (for US only): BIRD, Thomas, Geoffrey, Colerick [GB/FR]; ZENECA Pharma, Centre de Recherches, Z.I. La Pompelle, Chemin de Vrilly, Boîte postale 1050, F-51689 Reims Cedex 2 (FR). BARLAAM, Bernard, Christophe [FR/FR]; ZENECA Pharma, Centre de Recherches, Z.I. La Pompelle, Chemin de Vrilly, Boîte postale 1050, F-51689 Reims Cedex 2 (FR). LAMBERT, Christine, Marie, Paul [BE/FR]; ZENECA Pharma, Centre de Recherches, Z.I. La Pompelle, Chemin de Vrilly, Boîte postale 1050, F-51689 Reims Cedex 2 (FR).

(74) Agent: DENERLEY, Paul, Millington; Zeneca Pharmaceuticals, Intellectual Property Dept., Mereside, Alderley Park, Macclesfield, Cheshire SK10 4TG (GB).

(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ARIPO patent (GH, KE, LS, MW, SD, SZ, UG), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG).

Published
With international search report.
Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(54) Title: THIO DERIVATIVES OF HYDROXAMIC ACIDS



(I)

(57) Abstract

Compounds of formula (I), wherein R^1 is an aryl, arylC_{1-6}alkyl, heteroaryl or heteroarylC_{1-6}alkyl group; R^2 is hydrogen, C_{1-8}alkyl, C_{2-6}alkenyl, C_{2-6}alkynyl, C_{3-8}cycloalkyl, heteroaryl, heterocyclyl, arylC_{1-6}alkyl, heteroarylC_{1-6}alkyl, heterocyclylC_{1-6}alkyl or C_{3-8}cycloalkylC_{1-6}alkyl; R^3 is C_{1-6}alkyl, C_{2-6}alkenyl, aryl, C_{1-6}alkyl, heteroarylC_{1-6}alkyl or the side-chain of a naturally occurring amino acid; R^4 is hydrogen, C_{1-6}alkyl, C_{3-8}cycloalkyl, C_{4-8}cycloalkenyl, arylC_{1-6}alkyl, heteroarylC_{1-6}alkyl or heterocyclylC_{1-6}alkyl; R^5 is hydrogen or C_{1-6}alkyl; or R^4 and R^5 together with the nitrogen atom to which they are joined form a heterocyclic ring; wherein any group or ring, in R^1-R^5, is optionally substituted; or pharmaceutically acceptable salts or *in vivo* hydrolysable esters thereof, are described as inhibitors of the production of Tumour Necrosis Factor and/or one or more matrix metalloproteinase enzymes. Compositions containing them and their preparation are also described.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece			TR	Turkey
BG	Bulgaria	HU	Hungary	ML	Mali	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MN	Mongolia	UA	Ukraine
BR	Brazil	IL	Israel	MR	Mauritania	UG	Uganda
BY	Belarus	IS	Iceland	MW	Malawi	US	United States of America
CA	Canada	IT	Italy	MX	Mexico	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NE	Niger	VN	Viet Nam
CG	Congo	KE	Kenya	NL	Netherlands	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NO	Norway	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	NZ	New Zealand		
CM	Cameroon			PL	Poland		
CN	China	KR	Republic of Korea	PT	Portugal		
CU	Cuba	KZ	Kazakstan	RO	Romania		
CZ	Czech Republic	LC	Saint Lucia	RU	Russian Federation		
DE	Germany	LI	Liechtenstein	SD	Sudan		
DK	Denmark	LK	Sri Lanka	SE	Sweden		
EE	Estonia	LR	Liberia	SG	Singapore		

THIO DERIVATIVES OF HYDROXAMIC ACIDS

This invention relates to thio compounds and in particular to thio compounds wherein a thio substituent is located adjacent to a hydroxycarbamate group. This invention further relates to processes for preparing such thio compounds, to pharmaceutical compositions containing them and to their use in methods of therapeutic treatment.

The compounds of this invention are inhibitors of the production of TNF (Tumour Necrosis Factor) which is believed to be formed by the cleavage of a pro-form, or larger precursor, by the enzyme pro-TNF Convertase. Applicants believe that the compounds of the present invention inhibit TNF production by mechanisms which include inhibition of pro-TNF Convertase. The term 'TNF' is used herein to refer to Tumour Necrosis Factor in general but, in particular, to TNFα.

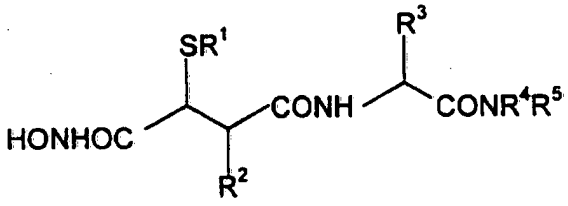
The compounds of this invention will be useful in the treatment of disease or medical conditions in which excessive TNF production is known to give rise via a cascade of processes to a variety of physiological sequelae including the production of physiologically-active eicosanoids such as the prostaglandins and leukotrienes, the stimulation of the release of proteolytic enzymes such as collagenase, the activation of osteoclast activity leading to the resorption of calcium, the stimulation of the release of proteoglycans from, for example, cartilage, the stimulation of cell proliferations and to angiogenesis. It is also known that, in certain cellular systems, TNF production precedes and mediates the production of other cytokines such as interleukin-1 (IL-1) and interleukin-2 (IL-2) which are also believed to contribute to the pathology of disease states such as inflammatory and allergic diseases and cytokine-induced toxicity. Excessive TNF production has also been implicated in mediating or exacerbating the development of various inflammatory and allergic diseases such as inflammation of the joints (especially rheumatoid arthritis, osteoarthritis and gout), inflammation of the gastrointestinal tract (especially inflammatory bowel disease, ulcerative colitis and gastritis), skin disease (especially psoriasis, eczema and dermatitis) and respiratory disease (especially asthma, bronchitis and allergic rhinitis), and in the production and development of various cardiovascular disorders such as myocardial infarction, angina and peripheral vascular disease. Excessive TNF production has also been implicated in mediating complications of bacterial, fungal and/or viral infections such as endotoxic shock, septic

shock and toxic shock syndrome. Excessive TNF production has also been implicated in mediating or exacerbating the development of adult respiratory distress syndrome, diseases involving cartilage or muscle resorption, Paget's disease and osteoporosis, pulmonary fibrosis, cirrhosis, renal fibrosis, the cachexia found in certain chronic diseases such as malignant disease and acquired immune deficiency syndrome (AIDS), tumour invasiveness and tumour metastasis and multiple sclerosis.

The compounds of the invention may also be inhibitors of one or more matrix metalloproteinases such as collagenases, stromelysins and gelatinases. Thus they may also be of use in the therapeutic treatment of disease conditions mediated by such enzymes for example arthritis (rheumatoid and osteoarthritis), osteoporosis and tumour metastasis.

The present invention provides novel thio compounds which have activity as inhibitors of TNF production and/or are inhibitors of one or more matrix metalloproteinase enzymes.

Accordingly the present invention provides a compound of the formula (I):



(I)

wherein:

R^1 is is aryl, arylC_{1-6}alkyl, heteroaryl or heteroarylC_{1-6} alkyl;

R^2 is is hydrogen, C_{1-6}alkyl, C_{2-6}alkenyl, C_{2-6}alkynyl, C_{3-8}cycloalkyl, aryl, heteroaryl, heterocyclyl, arylC_{1-6}alkyl, heteroarylC_{1-6}alkyl, heterocyclylC_{1-6}alkyl or C_{3-8}cycloalkylC_{1-6}alkyl;

R^3 is C_{1-6}alkyl, C_{2-6}alkenyl, arylC_{1-6}alkyl, heteroarylC_{1-6}alkyl or the side-chain of a naturally occurring amino acid;

R^4 is hydrogen, C_{1-6}alkyl, C_{3-8}cycloalkyl, C_{4-8}cycloalkenyl, arylC_{1-6}alkyl, heteroarylC_{1-6}alkyl or heterocyclylC_{1-6}alkyl;

R^5 is hydrogen or C_{1-6}alkyl; or R^4 or R^5 together with the nitrogen atom to which they are joined form a heterocyclic ring;

wherein any group or ring, in R^1-R^5, is optionally substituted;

or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof.

"Aryl in the terms "aryl" and "arylC_{1-6}alkyl" typically means phenyl or naphthyl, preferably phenyl. "Heteroaryl" in the terms "heteroaryl" and "heteroarylC_{1-6}alkyl" means an aromatic mono- or bicyclic 5-10 membered ring with up to five ring heteroatoms selected from nitrogen, oxygen and sulphur. Examples of 'heteroaryl' include thienyl, pyrrolyl, furanyl, imidazolyl, thiazolyl, pyrimidinyl, pyridinyl, indolyl, benzimidazolyl, benzthiazolyl, quinolinyl and isoquinolinyl. "Heterocyclyl" in the terms "heterocyclyl" and heterocyclyl-C_{1-6}alkyl means a non-aromatic mono- or bicyclic 5-10 membered ring with up to five ring hetero atoms selected from nitrogen, oxygen and sulphur. Examples of 'heterocyclyl' include pyrrolidinyl, morpholinyl, piperidinyl, dihydropyridinyl and dihydropyrimidinyl.

Any group or ring in R^1-R^5 may be optionally substituted, for example by up to three substituents which may be the same or different. Typical substituents include: hydroxy, C_{1-6}alkoxy for example methoxy, mercapto, C_{1-6}alkylthio for example methylthio, amino, C_{1-6}alkylamino for example methylamino, di-(C_{1-6}alkyl)amino for example dimethylamino, carboxy, carbamoyl, C_{1-6}alkylcarbamoyl for example methylcarbamoyl, di-C_{1-6}alkylcarbamoyl for example dimethylcarbamoyl, C_{1-6}alkylsulphonyl for example methylsulphonyl, arylsulphonyl for example phenylsulphonyl, C_{1-6}alkylaminosulphonyl for example methylaminosulphonyl, di-(C_{1-6}alkyl)aminosulphonyl for example dimethylamino-sulphonyl, nitro, cyano, cyanoC_{1-6}alkyl for example cyanomethyl, hydroxyC_{1-6}alkyl for example hydroxymethyl, aminoC_{1-6}alkyl for example aminoethyl, C_{1-6}alkanoylamino for example acetamido, C_{1-6}alkoxycarbonylamino for example methoxycarbonylamino, C_{1-6}alkanoyl for example acetyl, C_{1-6}alkanoyloxy for example acetoxy, C_{1-6}alkyl for example methyl, ethyl, isopropyl or tert-butyl, halo for example fluoro, chloro or bromo, trifluoromethyl, aryl for example phenyl, arylC_{1-6}alkyl for example benzyl, aryloxy for example phenoxy, arylC_{1-6}alkoxy for example benzyloxy, heteroaryl, heteroarylC_{1-6}alkyl, heterocyclyl and heterocyclylC_{1-6}alkyl. The term "side chain of a naturally occurring amino acid" means the side chain X of an amino acid NH_2-CHX-COOH. Suitable amino acids include alanine, arginine, aspartic acid, cysteine, asparagine, glutamine, histidine, homoserine, isoleucine, leucine, lysine, methionine, norleucine, norvaline, ornithine, serine, threonine, tryptophan, tyrosine and valine.

The compounds of the present invention possess a number of chiral centres, at -CH(SR^1)-, at -CHR^3-, at -CHR^2- (when R^2 is not hydrogen) and possibly in the variables R^1-R^5. The present invention covers all diastereoisomers and mixtures thereof that inhibit pro-TNF Convertase and/or inhibit matrix metalloproteinase enzymes.

In one aspect R^1 is an optionally substituted aryl group. Suitably R^1 is an optionally substituted phenyl group. In another aspect R^1 is optionally substituted arylC_{1-6}alkyl. In yet a further aspect R^1 is optionally substituted heteroaryl or heteroaryl-C_{1-6}alkyl.

Favourably R^1 is phenyl, phenylC_{1-6}alkyl, naphthylC_{1-6}alkyl, heteroaryl or heteroarylC_{1-6}alkyl wherein any of such rings is unsubstituted or substituted by one or two groups selected from halogen for example chloro or fluoro, C_{1-6}alkylcarbonyl for example acetyl, C_{1-6}alkylsulphonyl for example methylsulphonyl, trifluoromethyl, cyano, C_{1-6}alkyl for example methyl, isopropyl or tert-butyl, C_{1-6}alkoxy for example methoxy, cyanoC_{1-6}alkyl for example 1-cyano-1-methylethyl, or two adjacent carbon atoms on a phenyl ring are linked to form a methylenedioxy (-OCH_2O-) group.

In another aspect R^1 is phenyl, phenylC_{1-6}alkyl, naphthylC_{1-6}alkyl, heteroaryl or heteroarylC_{1-6}alkyl wherein any of such rings is unsubstituted or substituted by one or two groups selected from halogen for example chloro or fluoro, C_{1-6}alkylcarbonyl for example acetyl, C_{1-6}alkylsulphonyl for example methylsulphonyl, trifluoromethyl, C_{1-6}alkyl for example methyl, isopropyl or tert-butyl, C_{1-6}alkoxy for example methoxy, cyanoC_{1-6}alkyl for example 1-cyano-1-methylethyl, or two adjacent carbon atoms on a phenyl ring are linked to form a methylenedioxy (-OCH_2O-) group.

Further examples within the meaning of R^1 as an optionally substituted aryl group include those wherein two adjacent carbon atoms on a phenyl ring are linked by -$(CH_2)_m$- wherein m is 3 or 4, by -NR^a-CO-$(CH_2)_n$- wherein R^a is hydrogen or C_{1-6}alkyl and n is 1 or 2, by -NR^a-COCH=CH-, -CO-NR^a-$(CH_2)_n$- or by -CONR^a-CH=CH-.

Preferably R^1 is phenyl, 4-fluorophenyl, 4-trifluoromethylphenyl, 3,5-difluorophenyl, 4-acetylphenyl, 4-cyanophenyl, 4-methylsulphonylphenyl, 4-(1-cyano-1-methylethyl)phenyl, 3,4-dimethoxyphenyl, 3,5-dichlorophenyl or 3,5-di-trifluoromethylphenyl. Preferably also R^1 is 3-(1-cyano-1-methylethyl)phenyl, naphth-1-yl, 3-hydroxynaphth-7-yl or 2-chloro-4-fluorophenyl.

In a particular aspect R^1 is phenyl, 4-fluorophenyl, 4-trifluoromethylphenyl, 3,5-difluoro-phenyl, 4-acetylphenyl, 4-methylsulphonylphenyl, 4-(1-cyano-1-methylethyl)phenyl, 3,4-dimethoxyphenyl or 3,5-dichlorophenyl.

In particular R^1 may also be benzyl, phenethyl, phenylprop-1-yl, 1-methyl-phenylmethyl (PhCHMe-), 1,1-dimethylphenylmethyl ($PhCMe_2$-), thiazolyl, benzthiazolyl, 4-methoxybenzyl, indolyl, benzimidazolyl, indolylmethyl, pyrimidinyl, quinolinyl for example quinolin-2-yl, quinolin-6-yl or quinolin-7-yl, isoquinolinyl, pyridinyl or quinolinylmethyl for example quinolin-8-ylmethyl. Of these benzthiazolyl, quinolin-2-yl, quinolin-8-ylmethyl and benzyl are preferred.

In a particular aspect R^1 may be benzyl, phenethyl, phenylprop-1-yl, 1-methyl-phenylmethyl (PhCHMe-), 1,1-dimethylphenylmethyl ($PhCMe_2$-), thiazolyl, benzthiazolyl, 4-methoxybenzyl, indolyl, benzimidazolyl or indolylmethyl.

Particularly also R^1 may be 1-methyl-2-oxo-quinolin-6-yl, 1-methyl-2-oxodihydro-quinolinyl, 1-methyl-2-oxotetrahydroquinolinyl, 2-methyl-1-oxodihydroisoquinolinyl or 2-methyl-1-oxotetrahydroisoquinolinyl; of these 1-methyl-2-oxotetrahydroquinolin-7-yl is preferred.

Particular groups for R^2 include C_{1-8}alkyl for example isopropyl, n-propyl, isobutyl, sec-butyl, n-butyl, tert-butyl, isopentyl, n-pentyl, hexyl, heptyl or octyl; C_{1-8}alkyl interrupted by an oxygen or sulphur atom for example methoxypropyl, ethoxyethyl, propoxymethyl, ethylthioethyl, methylthiopropyl; phenylC_{1-6}alkyl for example benzyl, phenethyl, phenylpropyl or phenylbutyl; phenylC_{1-6}alkyl wherein the alkyl chain is interrupted by oxygen or sulphur for example benzyloxypropyl and benzyloxybutyl; C_{3-8}cycloalkyl for example cyclopropyl, cyclobutyl, cyclopentyl or cyclohexyl; or C_{3-8}cycloalkylC_{1-6}alkyl for example cyclopropylmethyl, cyclopropylethyl, cyclobutylmethyl, cyclopentylmethyl or cyclohexylmethyl.

In a particular aspect R^2 may be C_{1-8}alkyl for example isopropyl, n-propyl, isobutyl, sec-butyl, n-butyl, tert-butyl, isopentyl, n-pentyl, hexyl, heptyl or octyl; C_{1-8}alkyl interrupted by an oxygen or sulphur atom for example methoxypropyl, ethoxyethyl, propoxymethyl, ethylthioethyl, methylthiopropyl; phenylC_{1-6}alkyl for example benzyl, phenethyl, phenylpropyl or phenylbutyl; C_{3-8}cycloalkyl for example cyclopropyl, cyclobutyl, cyclopentyl

or cyclohexyl; or C_{3-8}cycloalkylC_{1-6}alkyl for example cyclopropylmethyl, cyclopropylethyl, cyclobutylmethyl, cyclopentylmethyl or cyclohexylmethyl.

Preferably R^2 is isobutyl.

There is a chiral centre at -CHR^2- (when R^2 is not hydrogen); it is preferred that this centre has the configuration indicated in formula (II) hereinafter. For most values of R^2 this centre will have the S-stereochemistry under the Cahn-Prelog-Ingold sequence rules.

Particular groups for R^3 include C_{1-6}alkyl for example methyl, ethyl, isopropyl, n-propyl, n-butyl, isobutyl, sec-butyl, tert-butyl, isopentyl, n-pentyl or hexyl; C_{1-6}alkyl interrupted by an oxygen or sulphur atom for example methoxyethyl, methoxypropyl, methylthioethyl or 1,1-dimethylmethylthiomethyl ($MeSCMe_2$-); or phenylC_{1-6}alkyl for example benzyl or phenethyl.

Preferably R^3 is isobutyl, tert-butyl, 1,1-dimethylmethylthiomethyl or benzyl with tert-butyl being most preferred.

The chiral centre at -CHR^3- preferably has the S-configuration indicated in formula (II) hereinafter. For most of R^3 this centre will have the S-stereochemistry.

Particular groups for R^4 include C_{1-6}alkyl for example methyl, ethyl, n-propyl, isopropyl, tert-butyl or n-butyl; C_{1-6}alkyl interrupted by an oxygen or sulphur atom for example hydroxyethyl, methoxyethyl, methylthioethyl or ethoxyethyl; phenylC_{1-6}alkyl for example benzyl, phenethyl or phenylpropyl; or C_{3-8}cycloalkylC_{1-6}alkyl for example cyclopropylmethyl, cyclobutylmethyl or cyclopentylmethyl.

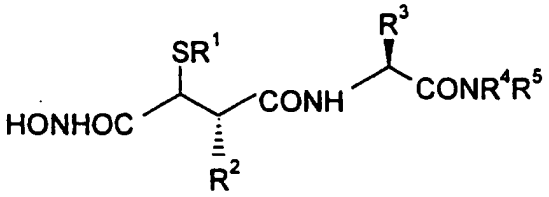
Particularly also, R^4 may be C_{1-6}alkylaminoC_{2-6}alkyl for example methylaminoethyl, di-C_{1-6}alkylaminoC_{2-6}alkyl for example dimethylaminoethyl or heterocyclicC_{1-6}alkyl for example 2-morpholinoethyl, 2-piperidinoethyl, 2-piperazinoethyl or 2-(N-methyl)piperazinoethyl.

Preferably R^4 is methyl, ethyl, n-propyl, isobutyl, tert-butyl, dimethylaminoethyl or benzyl. In one aspect R^4 is methyl, ethyl, n-propyl, isobutyl, tert-butyl or benzyl. Of these methyl is most preferred.

Particular groups for R^5 are hydrogen and C_{1-6}alkyl for example methyl or ethyl. Preferably R^5 is hydrogen.

In another aspect R^4 and R^5 together with the nitrogen atom to which they are joined form a heterocyclic ring, for example a 5 or 6 membered heterocyclic ring such as morpholino, piperidino, piperazino or N-methylpiperazino. Of these morpholino is preferred.

A particularly suitable class of compounds of the present invention is that of formula (II):



(II)

wherein R^1, R^2, R^3, R^4 and R^5 are as hereinbefore defined.

A preferred class of compounds of the formula (II) is that wherein R^1 is phenyl unsubstituted or substituted by one or two groups selected from halogen for example chloro or fluoro; C_{1-6}alkylsulphonyl for example methylsulphonyl; trifluoromethyl; C_{1-6}alkyl for example methyl, isopropyl or tert-butyl; C_{1-6}alkoxy for example methoxy; cyano; C_{1-6}alkanoyl for example acetyl; cyanoC_{1-6}alkyl for example 1-cyano-1-methylethyl; or two adjacent carbon atoms on the phenyl ring are linked to form a methylenedioxy (-OCH_2O-) group; R^2 is isobutyl; R^3 is isobutyl, tert-butyl, 1,1-dimethylmethylthiomethyl or benzyl; R^4 is methyl, ethyl, n-propyl, isobutyl, tert-butyl, dimethylaminoethyl, 2-morpholinoethyl or benzyl; and R^5 is hydrogen or methyl, or R^4 and R^5 together with the nitrogen atom to which they are joined form a morpholine ring.

In one aspect a preferred class of compounds of the formula (II) is that wherein R^1 is phenyl unsubstituted or substituted by one or two groups selected from halogen for example chloro or fluoro, C_{1-6}alkylsulphonyl for example methylsulphonyl, trifluoromethyl, C_{1-6}alkyl for example methyl, isopropyl or tert-butyl, C_{1-6}alkoxy for example methoxy, cyanoC_{1-6}alkyl for example 1-cyano-1-methylethyl, or two adjacent carbon atoms on the phenyl ring are linked to form a methylenedioxy (-OCH_2O-) group; R^2 is isobutyl; R^3 is isobutyl, tert-butyl or benzyl; R^4 is methyl, ethyl, n-propyl, isobutyl, tert-butyl or benzyl; and R^5 is hydrogen.

A further preferred class of compounds of the formula (II) is that wherein R^1 is phenylC_{1-6}alkyl, heteroaryl or heteroarylC_{1-6}alkyl wherein any of such rings is unsubstituted or substituted by one or two groups selected from halogen for example chloro or fluoro, C_{1-6}alkylsulphonyl for example methylsulphonyl, trifluoromethyl, C_{1-6}alkyl for example methyl, isopropyl or tert-butyl, C_{1-6}alkoxy for example methoxy, cyano, C_{1-6}alkanoyl for example acetyl, cyanoC_{1-6}alkyl for example 1-cyano-1-methylethyl, or two adjacent carbon atoms on the phenyl ring are linked to form a methylenedioxy ($-OCH_2O-$) group; R^2 is isobutyl; R^3 is isobutyl, tert-butyl, 1,1-dimethylmethylthiomethyl or benzyl; R^4 is methyl, ethyl, n-propyl, isobutyl, tert-butyl, dimethylaminoethyl, 2-morpholinoethyl or benzyl; and R^5 is hydrogen or methyl, or R^4 and R^5 together with the nitrogen atom to which they are joined form a morpholine ring.

In another aspect a class of compounds of the formula (II) is that wherein R^1 is phenylC_{1-6}alkyl, heteroaryl or heteroarylC_{1-6}alkyl wherein any of such rings is unsubstituted or substituted by one or two groups selected from halogen for example chloro or fluoro, C_{1-6}alkylsulphonyl for example methylsulphonyl, trifluoromethyl, C_{1-6}alkyl for example methyl, isopropyl or tert-butyl, C_{1-6}alkoxy for example methoxy, cyanoC_{1-6}alkyl for example 1-cyano-1-methylethyl, or two adjacent carbon atoms on the phenyl ring are linked to form a methylenedioxy ($-OCH_2O-$) group; R^2 is isobutyl; R^3 is isobutyl, tert-butyl or benzyl; R^4 is methyl, ethyl, n-propyl, isobutyl, tert-butyl or benzyl; and R^5 is hydrogen.

In yet another aspect, a preferred class of compounds is that wherein R^1 is quinolinyl, isoquinolinyl, 1-methyl-2-oxodihydroquinolinyl, 1-methyl-2-oxotetrahydroquinolinyl, 2-methyl-1-oxodihydroisoquinolinyl or 2-methyl-1-oxtetrahydroisoquinolinyl; R^2 is isobutyl; R^3 is isobutyl, tert-butyl, 1,1-dimethylmethylthiomethyl or benzyl; R^4 is methyl, ethyl, n-propyl, isobutyl, tert-butyl, dimethylaminoethyl, 2-morpholinoethyl or benzyl; and R^5 is hydrogen or methyl, or R^4 and R^5 together with the nitrogen atom to which they are joined form a morpholine ring.

Suitable pharmaceutically acceptable salts include acid addition salts such as hydrochloride, hydrobromide, citrate and maleate salts and salts formed with phosphoric and sulphuric acid. In another aspect suitable salts are base salts such as an alkali metal salt for example sodium or potassium, an alkaline earth metal salt for example calcium or magnesium, or organic amine salt for example triethylamine.

In vivo hydrolysable esters are those pharmaceutically acceptable esters that hydrolyse in the human body to produce the parent compound. Such esters can be identified by administering, for example intravenously to a test animal, the compound under test and subsequently examining the test animal's body fluids. Suitable in vivo hydrolysable esters for carboxy include methoxymethyl and for hydroxy include acetyl.

In order to use a compound of the formula (I) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof for the therapeutic treatment (including prophylactic treatment) of mammals including humans, it is normally formulated in accordance with standard pharmaceutical practice as a pharmaceutical composition.

Therefore in another aspect the present invention provides a pharmaceutical composition which comprises a compound of the formula (I) or a pharmaceutically acceptable salt or an in vivo hydrolysable ester and pharmaceutically acceptable carrier.

The pharmaceutical compositions of this invention may be administered in standard manner for the disease condition that it is desired to treat, for example by oral, topical, parenteral, buccal, nasal, vaginal or rectal administration or by inhalation. For these purposes the compounds of this invention may be formulated by means known in the art into the form of, for example, tablets, capsules, aqueous or oily solutions, suspensions, emulsions, creams, ointments, gels, nasal sprays, suppositories, finely divided powders or aerosols for inhalation, and for parenteral use (including intravenous, intramuscular or infusion) sterile aqueous or oily solutions or suspensions or sterile emulsions.

In addition to the compounds of the present invention the pharmaceutical composition of this invention may also contain, or be co-administered (simultaneously or sequentially) with, one or more pharmacological agents of value in treating one or more disease conditions referred to hereinabove.

The pharmaceutical compositions of this invention will normally be administered to humans so that, for example, a daily dose of 0.5 to 75 mg/kg body weight (and preferably of 0.5 to 30 mg/kg body weight) is received. This daily dose may be given in divided doses as necessary, the precise amount of the compound received and the route of administration depending on the weight, age and sex of the patient being treated and on the particular disease condition being treated according to principles known in the art.

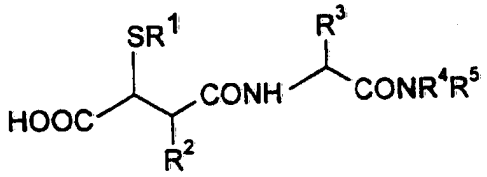
Typically unit dosage forms will contain about 1 mg to 500 mg of a compound of this invention.

Therefore in a further aspect, the present invention provides a compound of the formula (I) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof for use in a method of therapeutic treatment of the human or animal body.

In yet a further aspect the present invention provides a method of treating a disease condition mediated by TNF which comprises administering to a warm-blooded animal an effective amount of a compound of the formula (I) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof. The present invention also provides the use of a compound of the formula (I) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof in the preparation of a medicament for use in a disease condition mediated by TNF.

In another aspect the present invention provides a process for preparing a compound of the formula (I) or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof which process comprises

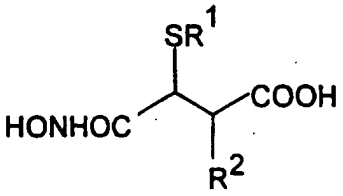
a) reacting a compound of the formula (III):



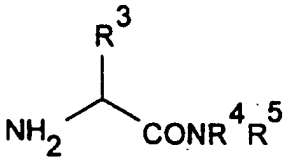
(III)

wherein R^1-R^5 are as hereinbefore defined, or an activated derivative thereof with hydroxylamine, O-protected hydroxylamine or a salt thereof; or

b) coupling a compound of the formula (IV) with a compound of the formula (V):



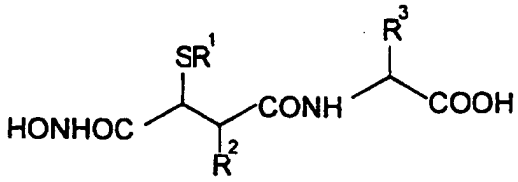
(IV)



(V)

wherein R^1-R^5 are as hereinbefore defined, under standard peptide coupling conditions; or

c) reacting a compound of the formula (VI) with compound of the formula (VII):



(VI)

HNR^4R^5 (VII)

wherein R^1-R^5 are as hereinbefore defined;

wherein any functional group is protected, if necessary, and:

i. removing any protecting groups;

ii. optionally forming a pharmaceutically acceptable salt or in vivo hydrolysable ester.

Protecting groups may in general be chosen from any of the groups described in the literature or known to the skilled chemist as appropriate for the protection of the group in question, and may be introduced by conventional methods.

Protecting groups may be removed by any convenient method as described in the literature or known to the skilled chemist as appropriate for the removal of the protecting group in question, such methods being chosen so as to effect removal of the protecting group

with minimum disturbance of groups elsewhere in the molecule.

Specific examples of protecting groups are given below for the sake of convenience, in which "lower" signifies that the group to which it is applied preferably has 1-4 carbon atoms. It will be understood that these examples are not exhaustive. Where specific examples of methods for the removal of protecting groups are given below these are similarly not exhaustive. The use of protecting groups and methods of deprotection not specifically mentioned is of course within the scope of the invention.

A carboxyl protecting group may be the residue of an ester-forming aliphatic or araliphatic alcohol or of an ester-forming silanol (the said alcohol or silanol preferably containing 1-20 carbon atoms).

Examples of carboxy protecting groups include straight or branched chain (1-12C)alkyl groups (eg isopropyl, t-butyl); lower alkoxy lower alkyl groups (eg methoxymethyl, ethoxymethyl, isobutoxymethyl); lower aliphatic acyloxy lower alkyl groups, (eg acetoxymethyl, propionyloxymethyl, butyryloxymethyl, pivaloyloxymethyl); lower alkoxycarbonyloxy lower alkyl groups (eg 1-methoxycarbonyloxyethyl, 1-ethoxycarbonyloxyethyl); aryl lower alkyl groups (eg benzyl, p-methoxybenzyl, o-nitrobenzyl, p-nitrobenzyl, benzhydryl and phthalidyl); tri(lower alkyl)silyl groups (eg trimethylsilyl and t-butyldimethylsilyl); tri(lower alkyl)silyl lower alkyl groups (eg trimethylsilylethyl); and (2-6C)alkenyl groups (eg allyl and vinylethyl).

Methods particularly appropriate for the removal of carboxyl protecting groups include for example acid-, base-, metal- or enzymically-catalysed hydrolysis.

Examples of hydroxyl protecting groups include lower alkyl groups (eg t-butyl), lower alkenyl groups (eg allyl); lower alkanoyl groups (eg acetyl); lower alkoxycarbonyl groups (eg t-butoxycarbonyl); lower alkenyloxycarbonyl groups (eg allyloxycarbonyl); aryl lower alkoxycarbonyl groups (eg benzoyloxycarbonyl, p-methoxybenzyloxycarbonyl, o-nitrobenzyloxycarbonyl, p-nitrobenzyloxycarbonyl); tri lower alkylsilyl (eg trimethylsilyl, t-butyldimethylsilyl) and aryl lower alkyl (eg benzyl) groups.

Examples of amino protecting groups include formyl, aralkyl groups (eg benzyl and substituted benzyl, p-methoxybenzyl, nitrobenzyl and 2,4-dimethoxybenzyl, and triphenylmethyl); di-p-anisylmethyl and furylmethyl groups; lower alkoxycarbonyl (eg

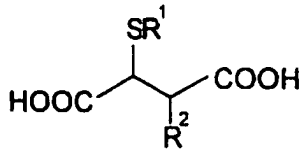
t-butoxycarbonyl); lower alkenyloxycarbonyl (eg allyloxycarbonyl); aryl lower alkoxycarbonyl groups (eg benzyloxycarbonyl, p-methoxybenzyloxycarbonyl, o-nitrobenzyloxycarbonyl, p-nitrobenzyloxycarbonyl); trialkylsilyl (eg trimethylsilyl and t-butyldimethylsilyl); alkylidene (eg methylidene); benzylidene and substituted benzylidene groups.

Methods appropriate for removal of hydroxy and amino protecting groups include, for example, acid-, base-, metal- or enzymically-catalysed hydrolysis, for groups such as p-nitrobenzyloxycarbonyl, hydrogenation and for groups such as o-nitrobenzyloxycarbonyl, photolytically.

The hydroxylamine group (HONH-), in particular in process variants (b) and (c), is typically O-protected for example with benzyl, 4-methoxybenzyl, 2,4-dimethoxybenzyl, t-butyl or a silyl (for example trimethylsilyl) group.

The compound of the formula (III) may be reacted in the form of the acid or an activated derivative thereof such as an acid halide, acid anhydride or an 'activated' ester such as 1H-benzo[1,2,3]triazol-1-yl, 1-hydroxy-benzo[1,2,3]triazole, pentafluorophenyl or 2,4,5-trichlorophenyl in the presence of a carbodiimide. The reaction of the compound of the formula (III) and hydroxylamine is performed under standard conditions. Typically the reaction of an activated ester of a compound of the formula (III) and hydroxylamine or O-protected hydroxylamine is performed in the presence of a base, for example 2,6-lutidine, (optionally in the presence of dimethylaminopyridine) or N-methylmorpholine in an anhydrous aprotic solvent, for example dimethylformamide, at a non-extreme temperature, for example in the region -30° to +25°, preferably about 0°C.

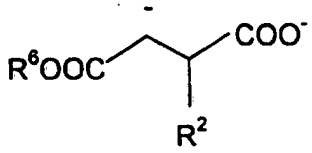
The compound of the formula (III) may be prepared by reacting a compound of the formula (VIII) with a compound of the formula (V):



(VIII)

wherein R^1 and R^2 are as hereinbefore defined, and wherein any functional group is protected and removed as necessary, under standard peptide coupling reaction conditions.

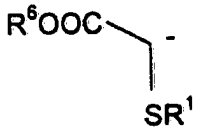
The compounds of the formula (VIII) may be prepared by reacting a source of -SR^1 with a dianion (formed in situ) of the formula (IX):



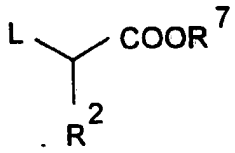
(IX)

wherein R^2 is as hereinbefore defined and R^6 is a protecting group, with subsequent removal of the protecting group if necessary. The dianion may be formed in situ by the action on the corresponding methylene compound, of a strong base, for example lithium di-isopropylamide, in an anhydrous substantially inert solvent, for example tetrahydrofuran, dimethylformamide, dimethylsulfoxide or a mixture thereof, at a low temperature such as -78°C. The corresponding methylene compounds are known in the art, for example see PCT Patent Application WO 94/21625. The source of -SR^1 may conveniently be a disulfide R^1-S-S-R^1 which is reacted in situ with the dianion of the formula (IX).

The compounds of the formula (VIII) may also be prepared by reacting a compound of the formula (X) with a compound of the formula (XI):



(X)



(XI)

wherein R^1, R^2 and R^6 are as hereinbefore defined, R^7 is a protecting group and L is a leaving group, for example a methanesulphonyloxy, trifluoromethanesulphonyloxy or p-toluenesulphonyloxy group. The anion of the formula (X) may be formed in situ by the action of a strong base on the corresponding methylene compound.

The compounds of the formulae (IV) and (V) are reacted under standard peptide coupling conditions wherein any functional group is protected as necessary. The compounds of the formula (IV) may be prepared by reacting hydroxylamine with a compound of the formula (VIII), both compounds protected as necessary under conditions similar to those described above for preparing compounds of the formula (III). The compounds of the formula (V) may be prepared under standard conditions for acylation of an amine.

The compounds of the formulae (VI) and (VIII) are reacted under standard conditions for acylation of an amine with any functional groups protected as necessary. The compounds of the formula (VI) may be prepared by methods similar to those described above for preparing compounds of the formula (IV).

The following biological test methods, data and Examples serve to illustrate the present invention.

Isolated Enzyme Assay

The ability of the compounds of this invention to inhibit proTNFα convertase enzyme is assessed in an isolated enzyme assay (termed "CON2"). Partially purified proTNFα convertase enzyme is obtained from the membranes of THP-1 cells as follows. 1.5-2.0x10^6 cells/ml THP-1 cells (initially cultured in RPMI 1640 medium + 10%(v/v) FCS, 10%(v/v) M1, 2mM L-glutamine 100IU/ml penicillin and 100μg/ml streptomycin) are induced in RPMI 1640 containing 1μg/ml LPS (E. coli O111:B4), 2mM Hydroxyurea, 50μg/ml silica and 1%(v/v) FCS at 37°C in a humidified (5%CO_2/95%air) incubator. After 16 hours the cells are harvested from a 5L induction by centrifugation at 640xg for 15 minutes. The cell pellets are washed once in RPMI 1640 without additive (1L per 2x10^{10} cells) and re-centrifuged at 640xg for 10 minutes. Cell pellets are resuspended in 10mM sodium phosphate buffer pH 7.4, containing 1mM $MgCl_2$, 30mM NaCl, 5μM PMSF, 0.02%(w/v) sodium azide (Buffer A) plus a few micrograms DNAase using 3 times the volume of buffer to packed cell pellets. A polytron homogeniser is used to lyse the cells by 5x5sec bursts with 1-2 minutes cooling between each burst. 30 ml of this homogenate is layered onto 10 mls of 41% (w/v) sucrose made in Buffer A and centrifuged at 150,000xg for 1 hour in a swing out rotor. The membrane is collected from the interphase, diluted by addition of 4 volumes Buffer A and centrifuged at 150,000xg for 20 minutes. The pellet is then resuspended in, Buffer A containing 1%(w/v) Triton X-100 to a concentration of 1mg/ml

and mixed for 1 hour at 4°C. The solubilised protein is recovered by centrifugation for 30 minutes at 100,000xg at 4°C. The supernatant is applied to a 25ml gelatin-sepharose 4B column equilibrated in 10mM Tris-HCl pH 8.0, 100mM NaCl, 0.1%(w/v) Triton X-100, 200μM PMSF, 0.02%(w/v) azide, 1μM $ZnCl_2$ (Buffer B). After loading the column is washed with Buffer B. The gelatin-sepharose flowthrough plus the first 10mls of the wash is then recycled overnight (1ml/min) on a 30ml wheatgerm-sepharose column previously equilibrated in 10mM Tris-HCl pH 8.0, 0.1%(w/v) Triton X-100, 200μM PMSF, 0.02%(w/v) azide, 1μM $ZnCl_2$ (Buffer C). After loading the column is washed in Buffer C and the enzyme is eluted with Buffer C containing 300mM N-Acetyl Glucosamine. The active enzyme fractions are applied to a 1ml Mono Q column equilibrated in Buffer C. After loading and washing with Buffer C, enzyme is eluted using a 0-500mM NaCl gradient in Buffer C. Active enzyme fractions are pooled and used as partially purified proTNFα convertase. In all cases the active fractions are assayed using the fluorogenic synthetic peptide substrate assay described below. This enzyme preparation cleaves 21kD soluble proTNFα at the correct cleavage site (Ala-Val) and enzyme activity is inhibited by matrix metalloprotease inhibitors (Gearing, A.J.H. etal., 1995, J Leukocyte Biol., 57,774-777). 4',5'-Dimethoxy-fluoresceinyl Ser.Pro.Leu.Ala.Gln.Ala.Val.Arg.Ser.Ser.Ser.Arg.Cys(4-(3-succinimid-1-yl)-fluorescein)-NH_2 the substrate is used to measure proTNFα convertase enzyme activity in CON2. It was synthesised as follows. The peptidic part of the substrate was assembled on Fmoc-NH-Rink-MBHA-polystyrene resin either manually or on an automated peptide synthesiser by standard methods involving the use of Fmoc-amino acids and O-benzotriazol-1-yl-N,N,N',N'-tetramethyluronium hexafluorophosphate (HBTU) as coupling agent with at least a 4- or 5-fold excess of Fmoc-amino acid and HBTU. Ser^1 and Pro^2 were double-coupled. The following side chain protection strategy was employed; $Ser^1(Bu^t)$, Gln^5(Trityl), $Arg^{8,12}$(Pmc or Pbf), $Ser^{9,10,11}$(Trityl), Cys^{13}(Trityl). Following assembly, the N-terminal Fmoc protecting group was removed by treating the Fmoc-peptidyl-resin with piperidine in DMF. The amino-peptidyl-resin so obtained was acylated by treatment for 1.5-2hr at 70°C with 1.5-2 equivalents of 4',5'-dimethoxy-fluorescein-4(5)-carboxylic acid (Khanna & Ullman, Anal Biochem, 108, 156-161, 1980) which had been preactivated with diisopropylcarbodiimide and 1-hydroxybenzotriazole in DMF. The dimethoxyfluoresceinyl-peptide was then simultaneously deprotected and cleaved from the resin by treatment with trifluoroacetic acid

containing 5% each of water and triethylsilane. The dimethoxyfluoresceinyl-peptide was isolated by evaporation, trituration with diethyl ether and filtration. The isolated peptide was reacted with 4-(N-maleimido)-fluorescein in DMF containing diisopropylethylamine, the product purified by RP-HPLC and finally isolated by freeze-drying from aqueous acetic acid. The product was characterised by MALDI-TOF MS and amino acid analysis.

Test compounds are serially diluted in assay buffer (50mM Tris HCl, pH 7.4 containing 0.1% (w/v) Triton X-100 and 2mM $CaCl_2$) and 50µl of each concentration is added to appropriate wells of a 96 well plate and 50µl assay buffer is added to substrate alone (n=6) and substrate +enzyme (n=6) control wells. ProTNFα convertase enzyme (25µl; 0.0144 units/ml in assay buffer) is added to all wells, except substrate alone controls which receive 25µl assay buffer. (NB: One unit of enzyme activity is defined as the convertase enzyme concentration which converts 1nMole substrate/hour).

Plates are incubated for 15 minutes at 26°C, prior to addition of 25µl substrate (40µM stock solution in assay buffer). Plates are then incubated at 26°C for 18 hours and read on a Fluoroskan II fluorometer (plates are also read at time 0 to obtain background values). In this test, generally, compounds are of interest if they have activity below 500nM. By way of example the compound of Example 1 gave a figure of 0.8 nM.

Assessment in human cell line (THF-2)

The ability of the compounds of this invention to inhibit TNFα production is assessed in THP-1 cells which are a human myelomonocytic cell line which synthesise and secrete TNFα when stimulated with lipopolysaccharide. THP-1 cells ($4x10^5$ cells in 160µl medium RPMI 1640 + bicarbonate, penicillin, streptomycin and glutamine) are incubated with 20µl of test compounds (triplicates) in DMSO or appropriate vehicle, in a 96 well tissue culture (TC) plate, for 30 min at 37°C in a humidified (5%CO_2/95%air) incubator, prior to addition of 20µl lipopolysaccharide (LPS) (E. Coli. 0111:B4 (Sigma); final concentration 50 µg/ml). Each assay includes controls of THP-1 cells incubated with medium alone (six wells/plate) or with a standard TNFα inhibitor. The plates are then incubated for 6 hours at 37°C (humidified incubator) after which time 100µl samples are removed from each well and transferred to a 96 well plate for storage at -70°C for subsequent analysis of TNFα concentration by ELISA. In this test, generally, compounds are of interest if they have activity below 10µM.

Assessment in whole blood assay

The ability of the compounds of this invention to inhibit TNFα production is also assessed in a human whole blood assay (HWBA). Human whole blood secretes TNFα when stimulated with LPS. This property of blood forms the basis of an assay which is used as a secondary test for compounds which profile as active in the THP-1 test. Heparinized (10Units/ml) human blood obtained from volunteers is diluted 1:5 with medium (RPMI1640 + bicarbonate, penicillin, streptomycin and glutamine) and incubated (160μl) with 20μl of test compound (triplicates), in DMSO or appropriate vehicle, for 30 min at 37°C in a humidified (5%CO_2/95%air) incubator, prior to addition of 20μl LPS (E. coli. 0111:B4; final concentration 10μg/ml). Each assay includes controls of diluted blood incubated with medium alone (6 wells/plate) or a known TNFα inhibitor as standard. The plates are then incubated for 6 hours at 37°C (humidified incubator), centrifuged (2000rpm for 10 min; 4°C), plasma harvested (50-100μl) and stored in 96 well plates at -70°C before subsequent analysis for TNFα concentration by ELISA. In this test, generally, compounds are of interest if they have activity below 50μM.

In vivo assessment

The ability of the compounds of this invention as *ex vivo* TNFα inhibitors is assessed in the rat. Briefly, groups of male Wistar Alderley Park (AP) rats (180-210g) are dosed with compound (6 rats) or drug vehicle (10 rats) by the appropriate route e.g. peroral (p.o.), intraperitoneal (i.p.) , subcutaneous (s.c.). Ninety minutes later rats are sacrificed using a rising concentration of CO_2 and bled out via the posterior vena cavae into 5 Units of sodium heparin/ml blood. Blood samples are immediately placed on ice and centrifuged at 2000 rpm for 10 min at 4°C and the harvested plasmas frozen at -20°C for subsequent assay of their effect on TNFα production by LPS-stimulated human blood. The rat plasma samples are thawed and 175μl of each sample are added to a set format pattern in a 96U well plate. Fifty μl of heparinized human blood is then added to each well, mixed and the plate is incubated for 30 min at 37°C (humidified incubator). LPS (25μl; final concentration10μg/ml) is added to the wells and incubation continued for a further 5.5 hours. Control wells are incubated with 25μl of medium alone. Plates are then centrifuged for 10 min at 2000 rpm and 200μl of the supernatants are transferred to a 96 well plate and frozen at -20°C for subsequent analysis of TNF concentration by ELISA.

Data analysis by dedicated software calculates for each compound/dose:

Percent inhibition of TNFα = $\frac{\text{Mean TNFα (Controls) - Mean TNFα (Treated) X 100}}{\text{Mean TNFα (Controls)}}$

Pharmacokinetic test

To evaluate the clearance properties of the compounds of this invention a sensitive ex vivo pharmacokinetic test is employed which utilises the CON2 assay to evaluate clearance rate.

This is a generic test which can be used to estimate the clearance rate of compounds across a range of species. Animals (eg. rats, marmosets) are dosed iv with a soluble formulation of compound and at subsequent time points (e.g. 5, 10, 15, 20, 30, 45, 60, 120 min) blood samples are taken from an appropriate vessel into 10U heparin. Plasma fractions are obtained following centrifugation and the plasma proteins precipitated with ethanol (70% final concentration). After 30 mins at 4°C the plasma proteins are sedimented by centrifugation and the supernatant fraction is evaporated to dryness using a Savant speed vac. The sediment is reconstituted in CON2 assay buffer and subsequently analysed for compound content using the TNF convertase assay (CON2). Briefly, a compound concentration-response curve is constructed for the compound undergoing evaluation. Serial dilutions of the reconstituted plasma extracts are assessed for activity and the amount of compound present in the original plasma sample is calculated using the concentration-response curve taking into account the total plasma dilution factor.

Test as anti-arthritic agent

Activity of a compound as an anti-arthritic is tested as follows. Acid soluble native type II collagen was shown by Trentham et al. [1] to be arthritogenic in rats; it caused polyarthritis when administered in Freunds incomplete adjuvant. This is now known as collagen-induced arthritis (CIA) and similar conditions can be induced in mice and primates. Recent studies have shown that anti-TNF monoclonal antibodies [2] and TNF receptor-IgG fusion proteins [3] ameliorate established CIA indicating that TNF plays a key role in the pathophysiology of CIA. Moreover, the remarkable efficacy reported for anti-TNF monoclonal antibodies in recent rheumatoid arthritis clinical trials indicates that TNF plays a

major role in this chronic inflammatory disease. Thus CIA in DBA/1 mice as described in references 2 and 3 is a tertiary model which can be used to demonstrate the anti-arthritic activity of a compound.

1. Trentham, D.E. et al., (1977) J. Exp. Med., 146, 857.

2. Williams, R.O. et al., (1992) Proc Natl Acad Sci, 89, 9784.

3. Williams, R.O. et al., (1995) Immunology, 84, 433.

In the examples:

(a) NMR spectra were taken at 400 MHz;

(b) DMF means dimethylformamide;

(c) Evaporation of solvents was carried out under reduced pressure;

(d) LDA means lithium di-isopropylamide;

(e) THF means tetrahydrofuran;

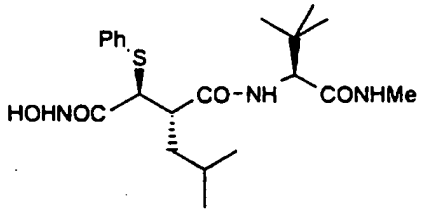
(f) DMSO means dimethylsulphoxide;

(g) AcOH means acetic acid.

(h) DMAP means dimethylaminopyridine.

Example 1

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide



N^2-[4-Hydroxy-2S-isobutyl-3S-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide (2.15 g, 5.1 mmol) was dissolved in DMF (15 ml). 1-Hydroxybenzotriazole (770 mg, 5.7 mmol) was added, followed by N-ethyl-N'-(3-dimethylaminopropyl)carbodiimide hydrochloride (1.09 g, 5.7 mmol). The mixture was stirred at room temperature for one hour. Hydroxylamine hydrochloride (529 mg, 7.6 mmol) was added immediately followed by 2,6-lutidine (830 µl, 7.6 mmol) and DMAP (62 mg, 0.51 mmol). The resulting solution was stirred at room temperature for 3 hours. The resulting mixture was purified by C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (gradient from 20/80 to 60/40). Elution yielded the title compound (520 mg; yield: 24%): m.p. = 198-200°C; ^{1}H-NMR (DMSO d-6): 0.76 (d, 3H, J= 6.6 Hz), 0.84 (d, 3H, J= 6.6 Hz), 0.93 (s, 9H), 0.9-1.0 (m, 1H), 1.3-1.5 (m, 2H), 2.55 (d, 3H, J= 4.8 Hz), 3.00 (m, 1H), 3.56 (d, 1H, J= 11.3 Hz), 4.22 (d, 1H, J= 9.5 Hz), 7.15-7.4 (m, 5H), 7.79 (q, 1H, J= 4.8 Hz), 8.00 (d, 1H, J= 9.5 Hz), 9.0 (s br, 1H), 10.75 (m, 1H); MS (ESI): 446 ($M + Na^+$).

N^2-[4-Hydroxy-2S-isobutyl-3S-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide used as the starting material was obtained as follows:

(i) To a stirred solution of LDA [32.8 mmol; prepared by addition of 1.6 M n-butyl lithium (20.5 ml, 32.8 mmol) in hexane to a solution of diisopropylamine (4.4 ml, 33.75 mmol) in dry THF (20 ml) at -78°C] cooled at -78°C under argon atmosphere was added 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester [(I)] (3.45 g, 15 mmol) in dry THF (15 ml) dropwise. The mixture was stirred for 90 minutes at -78°C and a solution of diphenyl disulfide (4.2 g, 19 mmol) in dry THF (15 ml) was added. The mixture was stirred for 30 minutes at -78°C, warmed to room temperature and stirred for two hours at room temperature. The solution was

cooled to -78°C and quenched by addition of methanol (6 ml). The solution was warmed to room temperature and the solvents were evaporated in vacuo. Ice was added to the residue and the mixture was acidified with 2N hydrochloric acid to pH 4. The solution was extracted with diethyl ether (3 x 100 ml). The combined organic extracts were dried over $MgSO_4$, filtered and the solvents were removed. The residue was purified by flash chromatography on silica using petroleum ether-ethyl acetate (gradient from 8/2 to 0/10) as eluant to give a mixture of 2S-isobutyl-3(R,S)-phenylthiobutan-1,4-dioic acid-4-tert-butyl ester (4.77g; ratio R/S: 50/50). Preparative normal phase HPLC purification using ethyl acetate-cyclohexane (5:95) as eluant gave 2S-isobutyl-3S-phenylthiobutan-1,4-dioic acid-4-tert-butyl ester (2.3 g): ^{1}H-NMR ($CDCl_3$): 0.83 (d, 3H, J= 7.3 Hz), 0.85 (d, 3H, J= 7Hz), 1.26 (m, 1H), 1.39 (s, 9H), 1.63 (m, 1H), 1.72 (m, 1H), 2.98 (m, 1H), 3.68 (d, 1H, J= 10.3 Hz), 7.28 (m, 3H), 7.50 (m, 2H).

Further elution gave the other isomer (2S-isobutyl-3R-phenylthiobutan-1,4-dioic acid-4-tert-butyl ester) (2.2 g): ^{1}H-NMR ($CDCl_3$): 0.93 (d, 6H, J= 6.2Hz), 1.35 (s, 9H), 1.6-1.75 (m, 2H), 1.86 (m, 1H), 2.81 (m, 1H), 3.62 (d, 1H, J= 10.3 Hz), 7.30 (m, 3H), 7.49 (m, 2H).

[1] British Biotech Ltd, (Crimmin, M.J.; Beckett, P.R.; Davis, M.H.), Patent WO94/21625 (1994)

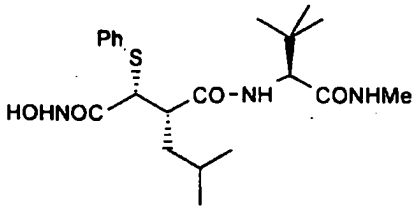
(ii) To 2S-isobutyl-3S-phenylthiobutan-1,4-dioic acid-4-tert-butyl ester (2.75 g, 8.1 mmol) in DMF (16 ml) at 0°C was added successively 1-hydroxybenzotriazole (1.32 g, 9.8 mmol) and N-ethyl-N'-(3-dimethylaminopropyl)carbodiimide hydrochloride (1.87 g, 9.8 mmol). After 15 minutes, L-tert-leucine methylamide (1.41 g, 9.8 mmol) was added to the mixture followed by DMAP (195 mg, 1.6 mmol). The mixture was stirred at room temperature for three hours. The mixture was poured into cold water and extracted with diethyl ether (2 x 100 ml). The combined organic layers were washed with saturated sodium bicarbonate, brine and dried over $MgSO_4$. The solvents were evaporated in vacuo and the residue was purified by flash chromatography on silica using ethyl acetate-petroleum ether (3:7) as eluant to give N^2-[2S-isobutyl-3S-phenylthio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide. (3.0 g, yield: 79%) as a white solid: ^{1}H-NMR ($CDCl_3$): 0.84 (d, 3H, J= 6.6 Hz), 0.90 (d, 3H, J= 6.2 Hz), 1.06 (s, 9H), 1.17 (m, 1H), 1.34 (s, 9H), 1.45 (m, 1H), 1.74 (m, 1H), 2.67 (m, 1H), 2.80 (d,

3H, J= 4.8 Hz), 3.71 (d, 1H, J= 11 Hz), 4.27 (d, 1H, J= 9.2 Hz), 5.95 (s br, 1H), 6.45 (d, 1H, J= 9.2 Hz), 7.27 (m, 3H), 7.47 (m, 2H).

(iii) Trifluoroacetic acid (8.3 ml) was added dropwise to a solution of N^2-[2S-isobutyl-3S-phenylthio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (3.0 g, 6.05 mmol) in dry dichloromethane (19 ml). The solution was stirred at 0°C overnight. The solvents were evaporated in vacuo. The residue was taken up in toluene and the solvent was removed in vacuo (three times) to give white crystals which were washed with pentane and dried in vacuo to yield N^2-[4-hydroxy-2S-isobutyl-3S-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide (2.56 g, 97%): ^{1}H-NMR (DMSO d-6): 0.79 (d, 1H, J= 6.6 Hz), 0.87 (d, 1H, J= 6.6 Hz), 0.96 (s, 9H), 1.06 (m, 1H), 1.40 (m, 1H), 1.56 (m, 1H), 2.57 (d, 3H, J= 4.4 Hz), 3.03 (m, 1H), 3.62 (d, 1H, J= 11 Hz), 4.26 (d, 1H, J= 9.2 Hz), 7.43-7.24 (m, 5H), 7.86 (q, 1H, J= 4.4 Hz), 8.13 (d br, 1H, J= 9.2 Hz).

Example 2

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3R-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide



N^2-[4-Hydroxy-2S-isobutyl-3R-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide (170 mg, 0.41 mmol) was dissolved in DMF (3.5 ml) and cooled at 0°C. 1-Hydroxybenzotriazole (73 mg, 0.54 mmol) was added, followed by N-methylmorpholine (60 µl, 0.54 mmol) and N-ethyl-N'-(3-dimethylaminopropyl)carbodiimide hydrochloride (103 mg, 0.54 mmol). The mixture was stirred at 0°C for one hour. A mixture of hydroxylamine hydrochloride (58 mg, 0.83 mmol) and N-methyl morpholine (91 µl, 0.83 mmol) in DMF (1.5 ml) was added. The resulting solution was stirred at 0°C for one hour then at room temperature overnight. The solvents were evaporated in vacuo and the residue was purified by C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (gradient from 20/80 to 60/40). Elution yielded the compound of Example 1 (55 mg; yield: 32%). Further elution using the

same solvent gave N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide (49 mg, yield: 29%): m.p.= 194-198°C; ^{1}H-NMR (DMSO d-6): 0.75 (d, 3H, J= 6.6 Hz), 0.79 (d, 3H, J= 6.6 Hz), 0.87 (s, 9H), 1.3-1.6 (m, 3H), 2.55 (d, 3H, J= 4.8 Hz), 2.88 (m, 1H), 3.62 (d, 1H, J= 9.5 Hz), 4.12 (d, 1H, J= 9.5 Hz), 7.2-7.5 (m, 5H), 7.69 (d, 1H, J= 9.5 Hz), 7.78 (q, 1H, J= 4.8 Hz), 8.86 (s, 1H), 10.75 (m, 1H); MS (ESI): 446 ($M + Na^+$).

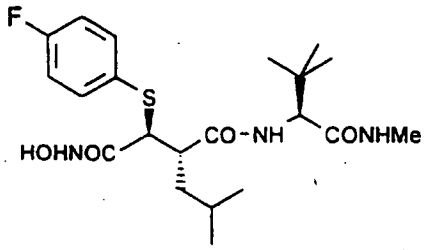
N^2-[4-Hydroxy-2S-isobutyl-3R-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide used as the starting material was obtained as follows:

(i) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3R-phenylthiobutan-1,4-dioic acid-4-tert-butyl ester (478 mg, 1.4 mmol) there was obtained N^2-[2S-isobutyl-3R-phenylthio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (540 mg, yield: 78%) as a white foam: ^{1}H-NMR ($CDCl_3$): 0.87 (d, 1H, J= 6.6 Hz), 0.88 (d, 1H, J= 6.6 Hz), 0.99 (s, 9H), 1.33 (s, 9H), 1.51 (m, 1H), 1.64 (m, 1H), 1.79 (m, 1H), 2.64 (m, 1H), 2.78 (d, 3H, J= 5.1 Hz), 3.73 (d, 1H, J= 9.9 Hz), 4.14 (d, 1H, J= 9.2 Hz), 5.88 (q, 1H, J= 5.1 Hz), 6.56 (d, 1H, J= 9.2 Hz), 7.30 (m, 3H), 7.49 (m, 2H).

(ii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3R-phenylthio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (490 mg) there was obtained N^2-[4-hydroxy-2S-isobutyl 3R-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide (460 mg) as a white solid: ^{1}H-NMR ($CDCl_3$): 0.84 (d, 3H, J= 6.2 Hz), 0.87 (d, 3H, J= 6.2 Hz), 1.06 (s, 9H), 1.55-1.80 (m, 3H), 2.86 (d, 3H, J= 5.1 Hz), 2.97 (m, 1H), 3.84 (d, 1H, J= 6.6 Hz), 4.38 (d, 1H; J= 9.5 Hz), 6.28 (s br, 1H), 7.32 (m, 3H), 7.50 (m, 2H).

Example 3

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(4-fluorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1, from N^2-[4-hydroxy-2S-isobutyl-3S-(4-fluorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (530 mg, 1.24 mmol) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(4-fluorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (240 mg, 36%) as a white solid: m.p.= 195-197°C; ^{1}H-NMR (DMSO d-6): 0.77 (d, 3H, J= 6.2 Hz), 0.85 (d, 3H, J= 6.6 Hz), 0.96 (s, 9H), 1.0 (m, 1H), 1.37 (m, 2H), 2.57 (d, 3H, J= 4.4 Hz), 3.01 (m, 1H), 3.46 (d, 1H, J= 11.4 Hz), 4.26 (d, 1H, J= 9.1 Hz), 7.17 (dd, 2H, J= J'= 8.8 Hz), 7.41 (dd, 2H, J=5.5 Hz, J'= 8.8 Hz), 7.81 (q, 1H, J= 4.4 Hz), 8.03 (d, 1H, J= 9.1 Hz), 8.98 (s br, 1H); 10.55 (s br, 1H); MS (ESI): 464 ($M + Na^+$).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i) , from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (1.15 g, 5 mmol) and di-(4-fluorophenyl) disulfide[2] (1.4 g), there was obtained 2S-isobutyl-3S-(4-fluorophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester: ^{1}H-NMR ($CDCl_3$): 0.90 (d, 1H, J= 6.6 Hz), 0.92 (d, 1H, J= 6.6 Hz), 1.25 (m, 1H), 1.41 (s, 9H), 1.75-1.55 (m, 2H), 2.94 (m, 1H), 3.56 (d, 1H, J= 10.6 Hz), 7.00 (dd, 2H, J=J'= 8.8 Hz), 7.50 (dd, 2H, J= 8.8 Hz, J'= 5.1 Hz) and the other isomer: 2S-isobutyl-3R-(4-fluorophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester: ^{1}H-NMR ($CDCl_3$): 0.93 (m, 6H), 1.37 (s, 9H), 1.9-1.3 (m, 3H), 2.77 (m, 1H), 3.54 (d, 1H, J= 10.3 Hz), 7.02 (dd, 2H, J=J'= 8.8 Hz), 7.49 (dd, 2H, J= 8.8 Hz, J'= 5.1 Hz).

(ii) In a manner analogous to that described in Example 1 (ii) except that dichloromethane was used instead of DMF as a solvent, from 2S-isobutyl-3S-(4-fluorophenyl)thiobutan-1,4-dioic

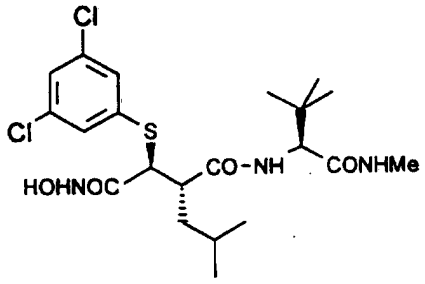
acid-4-tert-butyl ester (800 mg, 2.24 mmol), there was obtained N^2-[2S-isobutyl-3S-(4-fluorophenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (810 mg, 75%) as a white solid: ^{1}H-NMR ($CDCl_3$): 0.84 (d, 3H, J= 6.6 Hz), 0.89 (d, 3H, J= 6.6 Hz), 1.06 (s, 9H), 1.15 (m, 1H), 1.37 (s, 9H), 1.45 (m, 1H), 1.72 (m, 1H), 2.65 (m, 1H), 2.81 (d, 3H, J= 4.6 Hz), 3.61 (d, 1H, J= 11 Hz), 4.27 (d, 1H, J= 9.2 Hz), 5.88 (s br, 1H), 6.44 (d, 1H, J= 9.2 Hz), 6.98 (dd, J=J'= 8.8 Hz), 7.46 (dd, 2H, J= 5.1 Hz, J'= 8.8 Hz).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(4-fluorophenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (800 mg, 1.66 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(4-fluorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (700 mg) as a white solid: ^{1}H-NMR (DMSO d-6): 0.78 (d, 1H, J=6.6 Hz), 0.87 (d, 1H, J= 6.6 Hz), 0.97 (s, 9H), 1.05 (m, 1H), 1.6-1.35 (m, 2H), 2.58 (d, 1H, J= 4.4 Hz), 3.03 (m, 1H), 3.54 (d, 1H, J= 11.4 Hz), 4.28 (d, 1H, J= 9.5 Hz), 7.18 (m, 2H), 7.47 (dd, 2H, J= 5.5 Hz, J'= 8.8 Hz), 7.88 (q, 1H, J= 4.4 Hz), 8.13 (d, 1H, J= 9.5 Hz).

[2] Unless stated, the disulfides are prepared from the corresponding commercially available thiols by oxidation with iodine.

Example 4

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(3,5-dichlorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1, from N^2-[4-hydroxy-2S-isobutyl-3S-(3,5-dichlorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (770 mg) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(3,5-dichlorophenyl)-

thiosuccinyl]-L-tert-leucine-N^1-methylamide (80 mg, 11%) as a white solid; m.p.= 199-202°C: ^{1}H-NMR (DMSO d-6): 0.78 (d, 3H, J= 6.2 Hz), 0.86 (d, 3H, J= 6.2 Hz), 0.91 (s, 9H), 1.01 (m, 1H), 1.37 (m, 2H), 2.56 (d, 3H, J= 4.4 Hz), 3.10 (m, 1H), 3.68 (d, 1H, J= 11 Hz), 4.24 (d, 1H, J= 9.5Hz), 7.42 (d, 2H, J= 1.8 Hz), 7.46 (t, 1H, J= 1.8 Hz), 7.85 (q, 1H, J= 4.4 Hz), 8.13 (d, 1H, J= 9.5 Hz), 9.1 (s br, 1H), 10.6 (m, 1H); MS (ESI): 518 (M{^{37}Cl, ^{37}Cl} + Na^+), 516 (M{^{35}Cl, ^{37}Cl} + Na^+), 514 (M{^{35}Cl, ^{35}Cl} + Na^+).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i), from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (1.5 g, 6.5 mmol) and di-(3,5-dichlorophenyl) disulfide[2] (2.54 g), there was obtained 2S-isobutyl-3S-(3,5-dichlorophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.0 g): ^{1}H-NMR ($CDCl_3$): 0.92 (d, 3H, J= 6.6 Hz), 0.94 (d, 3H, J= 6.6 Hz), 1.25 (m, 1H), 1.43 (s, 9H), 1.8-1.5 (m, 2H), 2.97 (m, 1H), 3.70 (d, 1H, J= 10.3 Hz), 7.26 (1H), 7.38 (d, 2H, J= 1.8 Hz) and the other isomer: 2S-isobutyl-3R-(3,5-dichlorophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (300 mg): ^{1}H-NMR ($CDCl_3$): 0.94 (m, 6H), 1.40 (s, 9H), 1.8 1.5 (m, 3H), 2.83 (m, 1H), 3.69 (d, 1H, J= 10.2 Hz), 7.28 (1H, t, J= 1.8 Hz), 7.39 (d, 2H, J= 1.8 Hz).

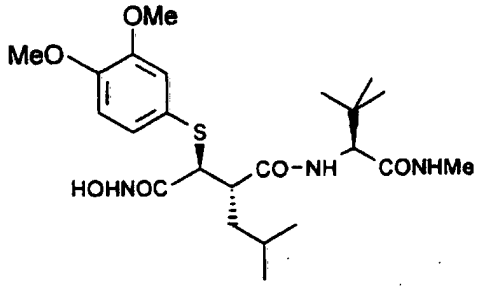
(ii) In a manner analogous to that described in Example 1 (ii) except that dichloromethane was used instead of DMF as a solvent, from 2S-isobutyl-3S-(3,5-dichlorophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1 g, 2.45 mmol), there was obtained N^2-[2S-isobutyl-3S-(3,5-dichlorophenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.0 g, 77%) as a white foam.

MS (ESI): 559 (M{^{37}Cl, ^{37}Cl} + Na^+), 557 (M{^{35}Cl, ^{37}Cl} + Na^+), 555 (M{^{35}Cl, ^{35}Cl} + Na^+)

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(3,5-dichlorophenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.0 g, 1.9 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(3,5-dichlorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (775 mg) as a white solid: ^{1}H-NMR (DMSO d-6): 0.80 (d, 3H, J= 6.6 Hz), 0.87 (d, 3H, J= 6.2 Hz), 0.93 (s, 9H), 1.1 (m, 1H), 1.40 (m, 1H), 1.57 (m, 1H), 2.56 (d, 3H, J= 4.4 Hz), 3.09 (m, 1H), 3.72 (d, 1H, J= 10.6 Hz), 4.26 (d, 1H, J=9.6 Hz), 7.45 (s, 2H), 7.52 (s, 1H), 7.90 (q, 1H, J= 4.4 Hz), 8.21 (d br, 1H, J= 9.6 Hz)

Example 5

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(3,4-dimethoxyphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1, from N^2-[4-hydroxy-2S-isobutyl-3S-(3,4-dimethoxyphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (450 mg) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(3,4-dimethoxyphenyl)-thiosuccinyl]-L-tert-leucine-N^1-methylamide (170 mg) as a white solid: m.p.= 218-220°C; ^{1}H-NMR (DMSO d-6): 0.77 (d, 3H, J= 6.2 Hz), 0.85 (d, 3H, J= 6.2 Hz), 0.98 (s, 9H), 1.0 (m, 1H), 1.40 (m, 2H), 2.58 (d, 3H, J= 4.4 Hz), 2.98 (m, 1H), 3.37 (d, 1H, J= 11.4 Hz), 3.75 (s, 3H), 3.76 (s, 3H), 4.29 (d, 1H, J= 9.5Hz), 6.88 (m, 2H), 6.99 (s, 1H), 7.81 (q, 1H, J= 4.4 Hz), 7.99 (d, 1H, J= 9.5 Hz), 8.98 (s br, 1H); 10.53 (s br, 1H); MS (ESI): 506 (M + Na^+).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i) , from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (1.0 g, 4.34 mmol) and di-(3,4-dimethoxyphenyl) disulfide[2] (2.0 g), there was obtained 2S-isobutyl-3S-(3,4-dimethoxyphenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.05 g): ^{1}H-NMR ($CDCl_3$): 0.90 (d, 3H, J= 6.6 Hz), 0.91 (d, 3H, J= 6.6 Hz), 1.26 (m, 1H), 1.42 (s, 9H), 1.75-1.55 (m, 2H), 2.96 (m, 1H), 3.54 (d, 1H, J= 10.3 Hz), 3.86 (s, 3H), 3.87 (s, 3H), 6.78 (d, 1H, J= 8.1 Hz), 7.1 (m, 2H) and the other isomer: 2S-isobutyl-3R-(3,4-dimethoxyphenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (550 mg): ^{1}H-NMR ($CDCl_3$): 0.94 (d, 6H, J= 6.6 Hz), 1.38 (s, 9H), 1.75-1.55 (m, 2H), 1.86 (m, 1H), 2.78 (m, 1H), 3.50 (d, 1H, J= 10.3 Hz), 3.87 (s, 3H), 3.88 (s, 3H), 6.80 (d, 1H, J= 8.1 Hz), 7.02 (d, 1H, J= 2.2 Hz), 7.09 (dd, 1H, J= 2.2 Hz, J'= 8.1 Hz).

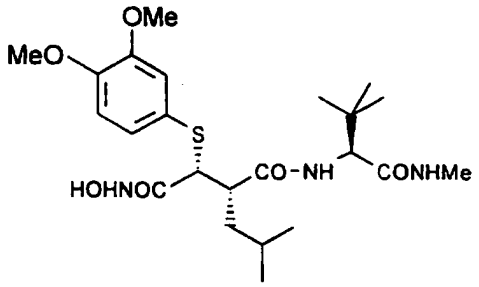
(ii) In a manner analogous to that described in Example 1 (ii) except that DMAP was replaced by 2,6-lutidine (1 equivalent), from 2S-isobutyl-3S-(3,4-dimethoxyphenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1 g, 2.45 mmol), there was obtained N^2-[2S-isobutyl-3S-(3,4-dimethoxyphenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.1 g, 84%) as a white foam.

MS (ESI): 547 ($M + Na^+$).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(3,4-dimethoxyphenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (900 mg, 1.7 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(3,4-dimethoxyphenyl)thio-succinyl]-L-tert-leucine-N^1-methylamide (795 mg) as a white solid: MS (ESI): 469 ($M + H^+$), 491($M + Na^+$).

Example 6

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3R-(3,4-dimethoxyphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1, from N^2-[4-hydroxy-2S-isobutyl-3R-(3,4-dimethoxyphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (450 mg) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(3,4-dimethoxyphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (170 mg) as a white solid. ^{1}H-NMR (DMSO d-6): 0.79 (d, 3H, J= 6.6 Hz), 0.82 (d, 3H, J= 6.2 Hz), 0.89 (s, 9H), 1.35-1.50 (m, 2H), 1.65 (m, 1H), 2.57 (d, 3H, J= 4.4 Hz), 2.82 (m, 1H), 3.45 (d, 1H, J= 9.9 Hz), 3.77 (s, 3H), 3.79 (s, 3H), 4.13 (d, 1H, J= 9.2Hz), 6.93 (d, 1H, J= 8.4 Hz), 7.04 (m, 2H), 7.73 (d, 1H, J= 9.2 Hz), 7.79 (s br, 1H), 8.86 (s, 1H); 10.65 (s br, 1H); MS (ESI): 506 ($M + Na^+$).

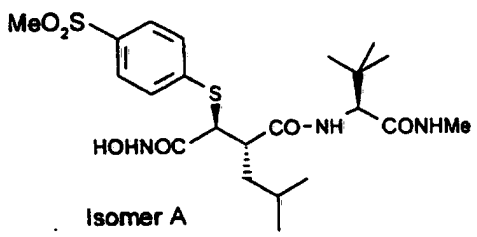
The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (ii) except that DMAP was replaced by 2,6-lutidine (1 equivalent), from 2S-isobutyl-3R-(3,4-dimethoxyphenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (480 mg, 1.2 mmol), there was obtained N^2-[2S-isobutyl-3R-(3,4-dimethoxyphenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (555 mg, 88%) as a white foam : MS (ESI): 547 ($M + Na^+$).

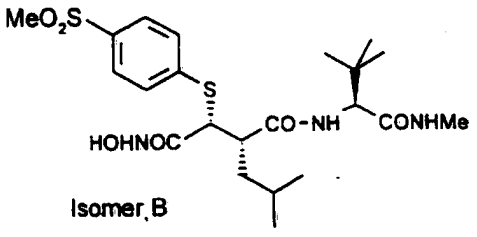
(ii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3R-(3,4-dimethoxyphenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (500 mg, 0.95 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3R-(3,4-dimethoxyphenyl)thio-succinyl]-L-tert-leucine-N^1-methylamide (256 mg) as a white solid.
MS (ESI): 469 ($M + H^+$), 491($M + Na^+$).

Example 7

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-S-(4-(methylsulfonyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer A) and
N^2-[4-(N-Hydroxyamino)-2S-isobutyl-R-(4-(methylsulfonyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer B)



Isomer A



Isomer B

In a manner analogous to that described in the first paragraph of Example 1 except that O-(trimethylsilyl)hydroxylamine was used instead of hydroxylamine hydrochloride, from the mixture of N^2-[4-hydroxy-2S-isobutyl 3(R/S)-(4-(methylsulfonyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (450 mg, 1:1 mixture) there was obtained N^2-[4-(N-hydroxyamino)

2S-isobutyl-3S-(4-(methylsulfonyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer A) (105 mg) as a white solid after purification of the crude mixture by C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (gradient from 0/100 to 45/55): m.p.= 222-224°C: ^{1}H-NMR (DMSO d-6): 0.83 (d, 3H, J= 6.6 Hz), 0.91 (d, 3H, J= 6.2 Hz), 0.94 (s, 9H), 1.08 (m, 1H), 1.44 (m, 1H), 1.55 (m, 1H), 2.60 (d, 3H, J= 4.8 Hz), 3.15 (m, 1H), 3.26 (s, 3H), 3.86 (d, 1H, J= 11.3 Hz), 4.23 (d, 1H, J= 9.2 Hz), 7.61 (d, 2H, J= 8.8 Hz), 7.83 (m, 3H), 8.1 (d, 1H, J= 9.2 Hz), 9.15 (s, 1H), 10.85 (s br, 1H); MS (ESI): 524 (M + Na$^+$).

Further elution with methanol and water/1%AcOH (45:55) gave N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(4-(methylsulfonyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer B) (77 mg) as a white solid: ^{1}H-NMR (DMSO d-6): 0.82 (d, 3H, J= 6.6 Hz), 0.85 (d, 3H, J= 6.6 Hz), 0.93 (s, 9H), 1.48 (m, 3H), 2.62 (d, 3H, J= 4.4 Hz), 3.03 (m, 1H), 3.27 (s, 3H), 3.41 (d, 1H, J= 9.2 Hz), 4.21 (d, 1H, J= 9.2 Hz), 7.71 (d, 2H, J= 8.4 Hz), 7.89 (m, 4H), 8.97 (s, 1H), 10.9 (s br, 1H); MS (ESI): 524 (M + Na$^+$).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i) except that di-(4-(methylsulfonyl)phenyl) disulfide was dissolved in DMF instead of THF, from 2R-isobutyl-butan-1,4-dioic acid-4-tert-butyl ester (1.31 g) and di-(4-(methylsulfonyl)phenyl) disulfide[3] (2.15 g), there was obtained a mixture of 2S-isobutyl-3(R/S)-(4-(methylsulfonyl)phenyl)thio-butan-1,4-dioic acid-4-tert-butyl ester (800 mg, 1:1 mixture): ^{1}H-NMR ($CDCl_3$): 0.94 (m, 6H), 1.38 (s, 9H, isomer B), 1.42 (s, 9H, isomer A), 1.8-1.3 (m, 3H), 2.89 (m, 1H, isomer B), 3.01 (m, 1H, isomer A), 3.05 (s, 3H), 3.85 (d, 1H, J= 10.2 Hz), 7.63 (m, 2H), 7.86 (m, 2H).

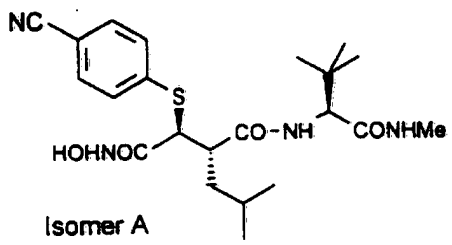
(ii) In a manner analogous to that described in Example 1 (ii) except that DMAP was replaced by 2,6 lutidine (1 equivalent), from the mixture of 2S-isobutyl-3(R/S)-(4-(methylsulfonyl)phenyl)thio-butan-1,4-dioic acid-4-tert-butyl ester (800 mg, 1:1 mixture), there was obtained a mixture of N^2-[2S-isobutyl 3(R/S)-(4-(methylsulfonyl)phenyl)thio 4-tert-butyloxy succinyl]-L-tert-leucine-N^1-methylamide (800 mg, 78%) as a white foam. MS (ESI): 565 (M + Na$^+$).

(iii) In a manner analogous to that described in Example 1 (iii), from the mixture of N^2-[2S-isobutyl 3(R/S)-4-(methylsulfonyl)phenylthio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (800 mg, 1.4 mmol, 1:1 mixture) there was obtained a mixture of N^2-[4-hydroxy-2S-isobutyl-3(R/S)-4-(methylsulfonyl)phenylthio succinyl]-L-tert-leucine-N^1-methylamide (500 mg) as a white solid: MS (ESI): 509 ($M + Na^+$).

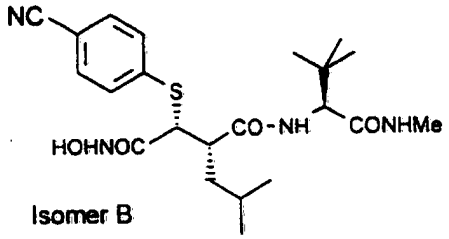
(3) Bordwell, F.G. et al.; J. Am. Chem. Soc., 1953, 75, 6019

Example 8

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-S-(4-cyanophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer A) and
N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3R-(4-cyanophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer B)



Isomer A



Isomer B

In a manner analogous to that described in the first paragraph of Example 1, from N^2-[4-hydroxy-2S-isobutyl-3S-(4-cyanophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (300 mg, 0.7 mmol) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(4-cyanophenyl)-thiosuccinyl]-L-tert-leucine-N^1-methylamide (68 mg, 22%) as a white solid (isomer A): m.p.= 238-241°C: ^{1}H-NMR (DMSO d-6): 0.78 (d, 3H, J= 6.6 Hz), 0.86 (d, 3H, J= 6.6 Hz), 0.88 (s, 3H); 0.97-1.08 (m, 1H), 1.3-1.53 (m, 2H), 2.55 (d, 3H, J= 4.8 Hz), 3.06-3.15 (m, 1H), 3.80 (d, 1H, J= 11 Hz), 4.17 (d, 1H, J= 9.2 Hz), 7.51 (d, 2H, J= 8.8 Hz), 7.73 (d, 2H, J= 8.5 Hz), 7.75-7.83 (m, 1H), 8.09 (d, 1H, J = 9.2 Hz), 9.05-9.15 (m,1H), 10.75-10.90 (m, 1H) and N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(4-cyanophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (20 mg) as a white solid (isomer B) formed by epimerisation during the reaction: ^{1}H-NMR

(DMSO d-6): 0.76 (d, 3H, J= 6 Hz), 0.80 (d, 3H, J = 6 Hz), 0.88 (s, 9H), 1.38-1.6 (m, 3H), 2.56 (d, 3H, J= 4.4 Hz), 2.97-3.04 (m, 1H), 3.88 (d, 1H, J= 9.5 Hz), 4.15 (d, 1H, J= 9.5 Hz), 7.60 (d, 2H, J= 8 Hz), 7.80 (d, 2H, J= 8.4 Hz), 7.75-7.85 (m, 2H), 8.94 (s, 1H), 10.85 (s, 1H).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i) , from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (1.5 g, 6.5 mmol) and di-(4-cyanophenyl) disulfide[4] (1.8 g), there was obtained 2S-isobutyl-3S-(4-cyanophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.2 g): ^{1}H-NMR ($CDCl_3$): 0.92 (m, 6H), 1.3 (m, 1H), 1.41(s, 9H), 1.8-1.55 (m, 2H), 2.99 (m, 1H), 3.82 (d, 1H, J= 9.9 Hz), 7.56 (m, 4H) and the other isomer: 2S-isobutyl-3R-(4-cyanophenyl)-thiobutan-1,4-dioic acid-4-tert-butyl ester (300 mg): ^{1}H-NMR ($CDCl_3$): 0.92 (d, 6H, J= 5.9 Hz), 1.37 (s, 9H), 1.8-1.5 (m, 3H), 2.89 (m, 1H), 3.82 (d, 1H, J= 10.2 Hz), 7.57 (m, 4H).

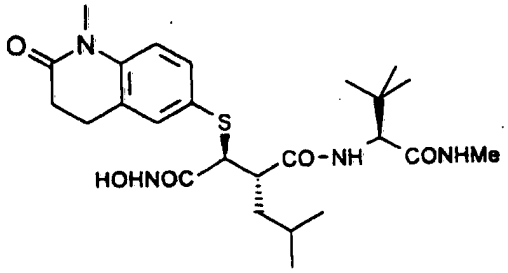
(ii) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3S-(4-cyanophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.1 g, 3 mmol), there was obtained N^2-[2S-isobutyl-3S-(4-cyanophenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.09 g, 74%) as a white foam: MS (ESI): 512 (M + Na^+).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(4-cyanophenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.09 g, 2.2 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(4-cyanophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (737 mg) as a white solid: MS (ESI): 434 (M + H^+), 456 (M + Na^+).

[4] Krishnamurthy, S. and al.; J. Org. Chem., 1989, 54, 4458

Example 9

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1, from N^2-[4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (360 mg, 0.73 mol) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (75 mg, 20%) as a white solid: m.p.= 235°C (decomposition): ^{1}H-NMR (DMSO d-6): 0.76 (d, 3H, J= 6.6 Hz), 0.85 (d, 3H, J= 6.2 Hz), 0.97 (s, 9H), 0.95-1.05 (m, 1H), 1.3-1.5 (m, 2H), 2.57 (d, 3H, J= 4.8 Hz), 2.5-2.6 (m, 2H), 2.83 (m, 2H), 3.00 (m, 1H), 3.24 (s, 3H), 3.46 (d, 1H, J= 11.3 Hz), 4.26 (d, 1H, J= 9.2 Hz), 7.02 (d, 1H, J= 8.5 Hz), 7.2 (s, 1H); 7.26 (d, 1H, J= 8.5 Hz), 7.8-7.85 (m, 1H), 8.00 (d, 1H, J= 9 Hz), 8.9-9.1 (m, 1H); 10.5-10.8 (m, 1H); MS (ESI): 529 ($M + Na^+$).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i) , from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (1.3 g, 5.65 mmol) and di-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl) disulfide[5] (2.8 g), there was obtained 2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiobutan-1,4-dioic acid-4-tert-butyl ester (950 mg) after purification on C18 preparative HPLC eluting with aqueous ammonium carbonate (0.2%)-acetonitrile (gradient from 100:0 to 72.5:27.5): MS (ESI): 444 ($M + Na^+$).

Further elution gave the other isomer: 2S-isobutyl-3R-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiobutan-1,4-dioic acid-4-tert-butyl ester (603 mg): MS (ESI): 444 ($M + Na^+$).

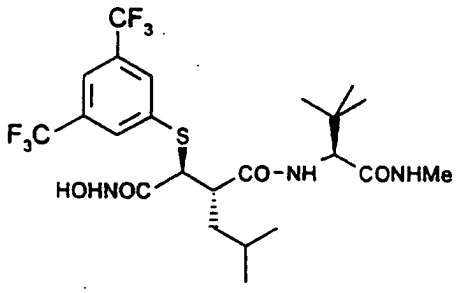
(ii) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl 3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiobutan-1,4-dioic acid 4-tert-butyl ester (390 mg, 0.92 mmol), there was obtained N^2-[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (250 mg,) as a white foam after C18 preparative HPLC eluting with aqueous ammonium carbonate (0.2%)-acetonitrile (gradient from 90:10 to 50:50): MS (ESI): 570 ($M + Na^+$).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (410 mg, 0.75 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (370 mg) as a white solid: ^{1}H-NMR ($CDCl_3$): 0.84 (d, 3H, J= 6.6 Hz), 0.88 (d, 3H, J= 6.3 Hz), 1.03 (s, 9H), 1.34 (m, 1H), 1.46 (m, 1H), 1.70 (m, 1H), 2.63 (m , 2H), 2.82 (d, 3H, J= 4.7 Hz), 2.86 (m, 2H), 2.93 (m, 1H), 3.81 (d, 1H, J= 9.2 Hz), 4.37 (d, 1H, J= 9.6 Hz), 6.38 (d, 1H, J= 8.4 Hz), 7.29 (d, 1H, J= 2.2 Hz), 7.40 (dd, 1H, J= 8.4 Hz, J'= 2.2 Hz), 7.7 (s br, 1H).

[5] Imperial Chemical Industries PLC, ICI-Pharma S.A.; (Bruneau, P.); EPA-462812

Example 10

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(3,5-di-(trifluoromethyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1, from N^2-[4-hydroxy-2S-isobutyl-3S-(3,5-di-(trifluoromethyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (500 mg, 0.91 mmol) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl 3S-(3,5-di-(trifluoromethyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (42 mg) as a white solid: m.p.= 225-230°C; ^{1}H-NMR (DMSO d-6): 0.78 (d, 3H, J= 6.6 Hz), 0.86 (s br, 12H), 1.0-1.1 (m, 1H), 1.3-1.5 (m, 2H), 2.56 (d, 3H, J= 4.8 Hz), 3.1-3.2 (m, 1H), 3.77 (d, 1H, J= 11.3 Hz), 4.25 (d, 1H, J= 9.5 Hz), 7.85-7.90 (m, 1H,), 7.94 (s, 1H), 8.04 (s, 2H), 8.15-8.21 (m, 1H), 9.0-9.12 (m, 1H), 10.8-10.92 (m, 1H); MS (ESI): 582 ($M + Na^+$).

The starting material was prepared as follows:

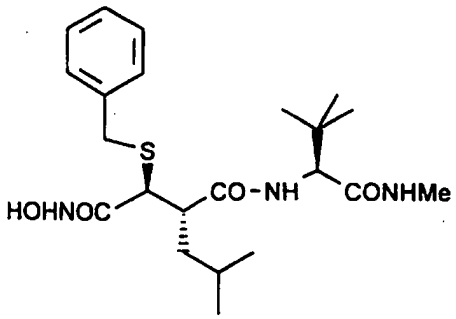
(i) In a manner analogous to that described in Example 1 (i) , from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (1.5 g, 6.5 mmol) and di-[3,5-di-(trifluoromethyl)phenyl] disulfide[2] (3.66 g), there was obtained 2S-isobutyl-3S-(3,5-di-(trifluoromethyl)phenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.3 g) after purification on C18 preparative HPLC eluting with methanol and water/1%AcOH (gradient from 20/80 to 70/30): MS (ESI): 497 ($M + Na^+$); the other isomer 2S-isobutyl-3R-(3,5-di-trifluoromethyl)phenylthiobutan-1,4-dioic acid 4-tert-butyl ester which was formed in the reaction as a minor component was not isolated.

(ii) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3S-(3,5-di-(trifluoromethyl)phenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.3 g, 2.7 mmol), there was obtained N^2-[2S-isobutyl-3S-(3,5-di-(trifluoromethyl)phenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.2 g) as a white foam: MS (ESI): 623 ($M + Na^+$).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(3,5-di-(trifluoromethyl)phenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.1g, 1.8 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(3,5-trifluoromethyl)phenyl)thio succinyl]-L-tert-leucine-N^1-methylamide (990 mg) as a white solid : MS (ESI): 545 ($M + H^+$), 567 ($M + Na^+$).

Example 11

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-benzylthiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1 except that O-(t-butyldimethylsilyl)hydroxylamine was used instead of hydroxylamine hydrochloride and no base was added, from N^2-[4-hydroxy-2S-isobutyl-3S-benzylthiosuccinyl]-L-tert-leucine-N^1-methylamide (150 mg, 0.35 mmol) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-benzylthiosuccinyl]-L-tert-leucine-N^1-methylamide (78 mg) as a white solid after addition of 1 ml of 1N hydrochloric acid to the crude mixture at the end of the reaction and purification of this mixture on C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (gradient from 0/100 to 60/40) m.p.= 200-205°C; ^{1}H-NMR (DMSO d-6): 0.75 (d, 3H, J= 6.2 Hz), 0.85 (d, 3H, J= 6.2 Hz), 0.95 (s, 9H), 1.00 (m, 1H), 1.45-1.30 (m, 2H), 2.55 (d, 3H, J= 4.4 Hz), 3.10 (m, 1H), 3.20 (d, 1H, J= 15 Hz), 3.83 (d, 1H, J= 12 Hz), 4.04 (d, 1H, J= 12 Hz), 4.26 (d, 1H, J= 9.5 Hz), 7.30-7.20 (m, 5H), 7.85 (d, 1H, J= 4.4 Hz), 8.05 (d, 1H, J= 9.5 Hz), 9.0 (s br, 1H), 10.64 (s br , 1H); MS (ESI): 460 ($M + Na^+$).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i) , from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (1.5 g, 6.5 mmol) and dibenzyl disulfide (3.66 g), there was obtained 2S-isobutyl-3R-benzylthiobutan-1,4-dioic acid-4-tert-butyl ester (1.48 g): ^{1}H-NMR ($CDCl_3$): 0.82 (d, 3H, J=6 Hz), 0.85 (d, 3H, J=5.8 Hz); 1.35 (m, 1H), 1.65-1.50 (m, 2H), 2.70 (m, 1H), 3.10 (d, 1H, J= 10 Hz), 3.84(d, 1H, J= 11.7 Hz), 3.87 (d, 1H, J= 11.7 Hz), 7.35-7.20 (m, 5H).

(ii) To a stirred solution of LDA [9.24 mmol; prepared by addition of 2.5 M n-butyl lithium (3.69 ml, 9.24 mmol) in hexane to a solution of diisopropylamine (1.29 ml, 9.24 mmol) in dry THF (5 ml) at -78°C] cooled at -78°C under an argon atmosphere was added 2S-isobutyl-3R-benzylthiobutan-1,4-dioic acid-4-tert-butyl ester (1.48 g, 4.20 mmol) in dry THF (10 ml) dropwise. The mixture was stirred for 15 minutes at -78°C, warmed to room temperature and stirred for two hours at room temperature. The solution was cooled to -78°C and quenched at -78°C by addition of methanol (3 ml). The solution was warmed to room temperature and the solvents were evaporated in vacuo. The residue was dissolved in dichloromethane and was washed successively with 1N hydrochloric acid and brine. The organic layer was dried over $MgSO_4$, filtered and the solvents were removed. The residue was purified by C18 preparative HPLC using methanol-aqueous 0.2% ammonium carbonate (60:40) as eluant. The fractions were collected, acidified to pH 2 with 2N hydrochloric acid and extracted with ether (2x100 ml). The organic layer was dried over $MgSO_4$ and evaporated to give 2S-isobutyl-3S-benzyl-thiobutan-1,4-dioic acid-4-tert-butyl ester (250 mg) as an oil: ^{1}H-NMR ($CDCl_3$): 0.84 (d, 3H, J= 6 Hz), 0.86 (d, 3H, J= 6 Hz), 1.20 (m, 1H), 1.50 (s, 9H), 1.65 (m, 2H), 2.92 (m, 1H), 3.16 (d, 1H, J= 10 Hz), 3.85 (s, 2H), 7.35-7.20 (m, 5H).

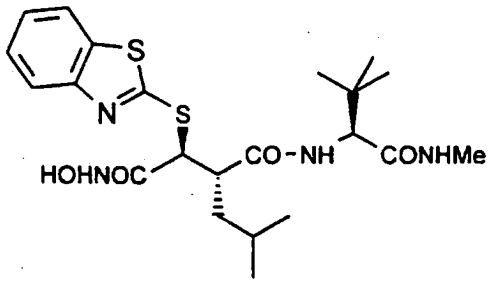
(iii) In a manner analogous to that described in Example 1 (ii) except that 2,6-lutidine (1 equivalent) was used instead of DMAP, from 2S-isobutyl-3S-benzylthiobutan-1,4-dioic acid-4-tert-butyl ester (225 mg, 0.64 mmol), there was obtained N^2-[2S-isobutyl-3S-benzylthio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (254 mg, 83 %) as a white foam.

MS (ESI): 501 ($M + Na^+$).

(iv) In a manner analogous to that described in example 1 (iii), from N^2-[2S-isobutyl-3S-benzylthio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (250 mg, 0.52 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-benzylthiosuccinyl]-L-tert-leucine-N^1-methyl-amide (165 mg) as a white solid: MS (ESI): 423 ($M + H^+$), 445 ($M + Na^+$).

Example 12

N^2-[4-(N-Hydroxyamino)-2S-isobutyl 3S-(benzothiazol-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1 except that O-(tert-butyldimethylsilyl)hydroxylamine (1.5 equivalents) was used instead of hydroxylamine hydrochloride and 1.3 equivalents of 2,6-lutidine was used, from N^2-[4-hydroxy-2S-isobutyl 3(R/S)-(benzothiazol-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (600 mg, 1.29 mmol) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(benzothiazol-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (125 mg) as a white solid after addition of 2 ml of 3N hydrochloric acid to the crude mixture at the end of the reaction and purification of this mixture on C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (gradient from 0/100 to 60/40): ^{1}H-NMR (DMSO d-6): 0.81 (d, 3H, J= 6.6 Hz), 0.87 (d, 3H, J= 6.6 Hz), 0.92 (s, 9H), 1.09 (m, 1H), 1.39 (m, 1H), 1.59 (m, 1H), 2.56 (d, 3H, J= 4.4 Hz), 3.20 (m, 1H), 4.17 (d, 1H, J= 9.9 Hz), 4.35 (d, 1H, J= 11 Hz), 7.39 (t, 1H, J= 7.7 Hz), 7.50 (t, 1H, J= 7.7 Hz), 7.81 (m, 1H), 7.89 (d, 1H, J= 7.7 Hz), 8.01 (d, 1H, J= 7.7 Hz), 8.09 (d, 1H, J=9.2Hz), 9.13 (s, 1H), 10.95 (s, 1H); MS (ESI): 481 ($M + H^+$), 503 ($M + Na^+$). Further elution gave N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(benzothiazol-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide.

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i) except that the solution of the dianion in THF cooled to -78°C was transferred through a double-ended needle into a slurry of di-(benzothiazol-2-yl) disulfide[2] in DMSO-THF (20 ml: 10 ml) cooled at 0°C and the mixture was stirred at 0°C for one hour, from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (1.15g,

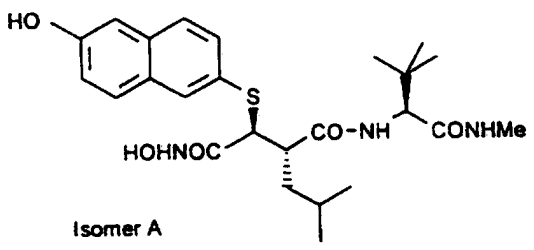
5 mmol) and di-(benzothiazol-2-yl) disulfide[2] (1.4 g), there was obtained a mixture of 2S-isobutyl-3(R/S)-(benzothiazol-2-yl)thiobutan-1,4-dioic acid-4-tert-butyl ester (830 mg, 1:1 mixture) after purification by C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (70/30): ^{1}H-NMR ($CDCl_3$): 0.96-0.87 (m, 6H), 1.41 and 1.42 (s, 9H), 1.9-1.6 (m, 3H), 3.20 and 3.29 (m, 1H), 4.77 (d, 1H, J= 8 Hz) and 4.79 (d, 1H, J= 7.6 Hz), 7.34 (m, 1H), 7.44 (m, 1H), 7.77 (m, 1H), 7.89 (m, 1H)

(ii) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3(R/S)-(benzothiazol-2-yl)thiobutan-1,4-dioic acid-4-tert-butyl ester (800 mg, 2.0 mmol), there was obtained N^2-[2S-isobutyl-3(R/S)-(benzothiazol-2-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (983 mg, 94%) as a white foam: MS (ESI): 522 (M + H^+), 544 (M + Na^+), 560 (M + K^+).

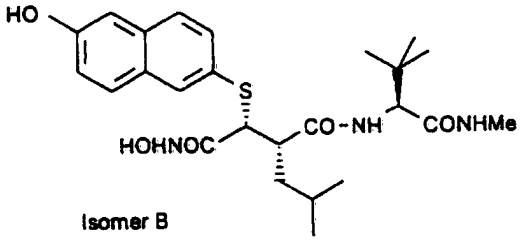
(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3(R/S)-(benzothiazol-2-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (900 mg, 1.72 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3(R/S)-(benzothiazol-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (725 mg, 91%) as a white solid: MS (ESI): 466 (M + H^+), 488 (M + Na^+).

Example 13

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(6-hydroxynaphth-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer A) and
N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3R-(6-hydroxynaphth-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer B)



Isomer A



Isomer B

In a manner analogous to that described in the first paragraph of Example 1, except that O-(t-butyldimethylsilyl)hydroxylamine was used instead of hydroxylamine hydrochloride and no base was added, from N^2-[4-hydroxy-2S-isobutyl-3S-(6-acetoxynaphth-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (650 mg, 1.26 mmol) there was obtained after treatment of the crude reaction mixture with HCl (2N, 10 drops) and purification by C18 preparative HPLC using as eluant a mixture of methanol and water/ 1% AcOH (gradient from 30/70 to 60/40) N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(6-hydroxynaphth-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer A) (65 mg, 10%) as a white solid: ^{1}H-NMR (DMSO d-6): 0.76 (d, 3H, J= 6.6 Hz), 0.84 (d, 3H, J= 6.6 Hz), 0.95 (s, 9H), 1.0 (m, 1H), 1.35-1.55 (m, 2H), 2.55 (d, 3H, J= 4.7 Hz), 3.05 (m, 1H), 3.56 (d, 1H, J= 11.4 Hz), 4.26 (d, 1H, J= 9.1 Hz), 7.1 (m, 2H), 7.36 (d, 1H, J= 8.4 Hz), 7.61 (d, 1H, J= 8.8 Hz), 7.68 (d, 1H J= 9.5 Hz), 7.77 (s, 1H), 7.8 (s br, 1H), 8.03 (d, 1H, J= 9.1 Hz), 8.93 (s br, 1H), 9.81 (s, 1H), 10.58 (s br, 1H); MS (ESI): 512 ($M + Na^+$).

Further elution with methanol and water/ 1% AcOH (60/40) gave N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(6-hydroxynaphth-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (Isomer B) (30 mg, 5%) as a white solid formed by epimerisation during the reaction: ^{1}H-NMR (DMSO d-6): 0.75 (d, 3H, J= 6.6 Hz), 0.80 (d, 3H, J= 6.6 Hz), 0.86 (s, 9H), 1.4 (m, 1H), 1.5 (m, 1H), 1.65 (m, 1H), 2.55 (d, 3H, J= 4.7 Hz), 2.88 (m, 1H), 3.61 (d, 1H, J= 9.5 Hz), 4.12 (d, 1H, J= 9.1 Hz), 7.1 (m, 2H), 7.45 (d, 1H, J= 8.8 Hz), 7.67 (d, 1H, J= 8.8 Hz), 7.74 (d, 1H, J= 9.9 Hz), 7.75 (m, 1H), 7.78 (m, 1H), 7.9 (s br, 1H), 8.85 (s br, 1H), 9.86 (s, 1H), 10.8 (s br, 1H); MS (ESI): 490 ($M + H^+$), 512 ($M + Na^+$).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i), from 2R-isobutylbutan-1,4-dioic acid 4-tert-butyl ester (1.92 g, 8.4 mmol) and di-(6-acetoxynaphth-2-yl) disulfide[6] (4.0 g), there was obtained 2S-isobutyl-3S-(6-acetoxynaphth-2-yl)thiobutan-1,4-dioic acid 4-tert-butyl ester (1.52g, 41%): ^{1}H-NMR ($CDCl_3$): 0.93 (d, 3H, J= 6.2 Hz), 0.94 (d, 3H, J= 6.2 Hz), 1.33 (s, 9H), 1.62-1.7 (m, 1H), 1.7-1.76 (m, 1H), 1.86-1.93 (m, 1H), 2.35 (s, 3H), 2.85 (m, 1H), 3.72 (d, 1H, J= 10.2 Hz), 7.23-7.26 (m, 1H), 7.53-7.56 (m, 2H), 7.73 (d, 1H, J= 8.4 Hz), 7.78 (d, 1H, J= 8.8 Hz), 7.99 (s br, 1H); MS (ESI): 469 ($M + Na^+$); the other isomer 2S-isobutyl-3R-(6-

acetoxynaphth-2-yl)thiobutan-1,4-dioic acid 4-tert-butyl ester which was formed in the reaction as a minor component was not isolated.

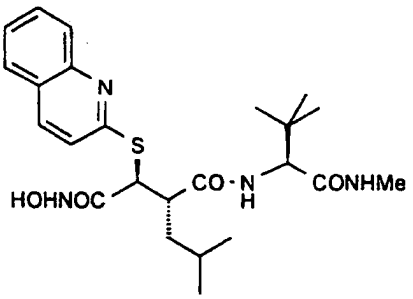
(ii) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3S-(6-acetoxynaphth-2-yl)thiobutan-1,4-dioic acid 4-tert-butyl ester (1.5 g, 3.36 mmol), there was obtained N^2-[2S-isobutyl-3S-(6-acetoxynaphth-2-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.26 g, 65%) as an off-white solid: ^{1}H-NMR ($CDCl_3$): 0.87 (d, 3H, J= 6.6 Hz), 0.89 (d, 3H, J= 6.6 Hz), 0.98 (s, 9H), 1.30 (s, 9H), 1.53 (m, 1H), 1.65 (m, 1H), 1.86 (m, 1H), 2.35 (s, 3H), 2.7 (m, 1H), 2.76 (d, 3H, J= 4.4 Hz), 3.79 (d, 1H, J= 10.2 Hz), 4.28 (d, 1H, J= 9.5 Hz), 6.58-6.66 (m, 2H), 7.25 (m, 1H), 7.53-7.57 (m, 2H), 7.73 (d, 1H, J= 8.4 Hz), 7.77 (d, 1H, J= 9.1 Hz), 7.99 (s, 1H); MS (ESI): 573 ($M + H^+$), 595 ($M + Na^+$).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(6-acetoxynaphth-2-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (780 mg, 1.36 mmol) there was obtained after purification by flash chromatography on silica using ethyl acetate-methanol (4:1) as eluant N^2-[4-hydroxy-2S-isobutyl-3S-(6-acetoxynaphth-2-yl)thio succinyl]-L-tert-leucine-N^1-methylamide (650 mg, 92%) as a white solid: ^{1}H-NMR (DMSO d-6): 0.78 (d, 3H, J= 5.8 Hz), 0.82 (d, 3H, J= 5.5 Hz), 0.92 (s, 9H), 1.55-1.65 (m, 3H), 2.32 (s, 3H), 2.62 (d, 3H, J= 4.4 Hz), 2.8 (m, 1H), 3.88 (d, 1H, J= 7.3 Hz), 4.03 (m, 1H), 7.3 (dd, 1H, J= 2.2 Hz, J'= 8.9 Hz), 7.56 (dd, 1H, J= 1.8 Hz, J'= 8.8 Hz), 7.63 (d, 1H, J= 2 Hz), 7.78 (m, 1H), 7.8-7.88 (m, 3H), 7.98 (s, 1H); MS (ESI): 539 ($M + Na^+$), 555 ($M + K^+$).

[6]Prepared by acetylation using the standard method (acetyl chloride, triethylamine in THF) from the corresponding commercially available disulfide.

Example 14

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(quinolin-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1 except that O-(t-butyldimethylsilyl)hydroxylamine (1.3 equivalent) was used instead of hydroxylamine hydrochloride, 2,6-lutidine (1.3 equivalent) and DMAP (0.1 equivalent) were used as a base, from N^2-[4-hydroxy-2S-isobutyl-3(R/S)-(quinolin-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (400 mg, 0.87 mmol) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(quinolin-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (135 mg) as a white solid after purification of the crude mixture by C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (gradient from 0/100 to 60/40): m.p.= 188-190°C; ^{1}H-NMR (DMSO d-6): 0.82 (d, 3H, J= 6.6 Hz), 0.88 (d, 3H, J= 6.6 Hz), 0.92 (s, 9H), 1.16 (m, 1H), 1.42 (m, 1H), 1.62 (m, 1H), 2.55 (d, 3H, J= 4.4 Hz), 3.19 (m, 1H), 4.14 (d, 1H, J= 9.2 Hz), 4.54 (d, 1H, J= 11.4 Hz), 7.30 (d, 1H, J= 8.8 Hz), 7.52 (t, 1H, J= 7.3 Hz), 7.76 (m, 2H), 7.90 (d, 1H, J= 8.8 Hz), 8.16-8.04 (m, 3H), 9.05 (s br, 1H), 10.75 (s br, 1H); MS (ESI): 475 ($M + H^+$), 497 ($M + Na^+$).

Further elution gave N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(quinolin-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide contaminated with minor impurities.

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i) , from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (1.1 g, 4.8 mmol) and di-(quinolin-2-yl) disulfide[2] (1.4 g), there was

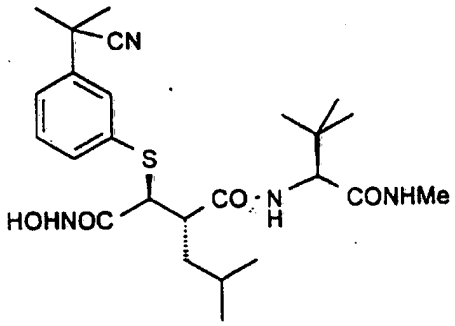
obtained 2S-isobutyl-3(R/S)-(quinolin-2-yl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.14 g, 68 %): MS (ESI): 412 (M + Na^+).

(ii) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3(R/S)-(quinolin-2-yl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.08 g, 2.8 mmol), there was obtained N^2-[2S-isobutyl-3(R/S)-(quinolin-2-yl)thio-4-tert-butyloxy-succinyl]-L-tert-leucine-N^1-methylamide (1.16 g, 80%, 4:6 mixture) as a white foam.

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3(R/S)-(quinolin-2-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (800 mg, 1.55 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3(R/S)-(quinolin-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (410 mg) as a white solid: MS (ESI): 482 (M + Na^+).

Example 15

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-[3-(1-cyano-1-methylethyl)phenyl]thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1 except that O-(t-butyldimethylsilyl)hydroxylamine (1.1 equivalents) was used instead of hydroxylamine hydrochloride and 2,6-lutidine (1 equivalent) was used as a base, from N^2-[4-hydroxy-2S-isobutyl-3S-[3-(1-cyano-1-methylethyl)phenyl]thiosuccinyl]-L-tert-leucine-N^1-methylamide (410 mg, 0.86 mmol) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-[3-(1-cyano-1-methylethyl)phenyl]thiosuccinyl]-L-tert-leucine-N^1-methylamide (170 mg, 40%) as a white solid after addition of 2 ml of 2N hydrochloric acid to the crude mixture at the end of

the reaction and purification of this mixture on C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (gradient from 20/80 to 65/35): ^{1}H-NMR (DMSO d-6): 0.78 (d, 3H, J= 6.6 Hz), 0.86 (d, 3H, J= 6.6 Hz), 0.94 (s, 9H), 1.01 (m, 1H), 1.37 (m, 1H), 1.47 (m, 1H), 1.69 (s, 6H), 2.57 (d, 3H, J= 4.4 Hz), 3.04 (m, 1H), 3.62 (d, 1H, J= 11.4 Hz), 4.25 (d, 1H, J= 9.2 Hz), 7.44-7.35 (m, 4H), 7.81 (m, 1H), 8.04 (d, 1H, J= 9.2 Hz), 9.05 (s, 1H), 10.72 (s, 1H); MS (ESI): 513 (M + Na^+).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i) , from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (0.91 g, 3.9 mmol) and di-[3-(1-cyano-1-methylethyl)phenyl] disulfide[2] (1.6 g, 4.5 mmol), there was obtained 2S-isobutyl-3S-[3-(1-cyano-1-methylethyl)-phenyl]thiobutan-1,4-dioic acid-4-tert-butyl ester (565 mg, 36 %): MS (ESI): 428 (M + Na^+). Further elution gave 2S-isobutyl-3R-[3-(1-cyano-1-methylethyl)phenyl]thiobutan-1,4-dioic acid-4-tert-butyl ester.

(ii) In a manner analogous to that described in Example 1 (ii) except that no DMAP was added, from 2S-isobutyl-3S-[3-(1-cyano-1-methylethyl)phenyl]thiobutan-1,4-dioic acid-4-tert-butyl ester (565 mg, 1.4 mmol), there was obtained N^2-[2S-isobutyl-3S-[3-(1-cyano-1-methylethyl)phenyl]thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (495 mg, 67%) as a white foam: MS (ESI): 554 (M + Na^+).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-[3-(1-cyano-1-methylethyl)phenyl]thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (492 mg, 0.93 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-[3-(1-cyano-1-methylethyl)phenyl]thiosuccinyl]-L-tert-leucine-N^1-methylamide (440 mg, 99%) as a white solid: MS (ESI): 498 (M + Na^+).

(iv) Di-[3-(1-cyano-1-methylethyl)phenyl] disulfide was prepared as follows:

1) alkylation of (3-bromophenyl)acetonitrile with excess sodium hydride/methyl iodide in THF gave 2-(3-bromophenyl) 2-methylpropionitrile (yield: 100%)

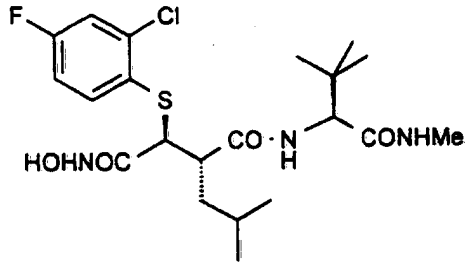
2) 2-(3-(tert-butylthio)phenyl) 2-methylpropionitrile was obtained from 2-(3-bromophenyl) 2-methylpropionitrile by reaction with tert-butyl mercaptan (1.3 eq.), potassium tert-butoxide(1.35 eq.) in DMSO at 80°C in the presence of $Pd(PPh_3)_4$ (0.05 eq) (yield: 37%)

3) 2-(3-mercaptophenyl) 2-methylpropionitrile was obtained from 2-(3-(tert-butylthio)phenyl) 2-methylpropionitrile by reaction with trifluoroacetic acid (1,7 eq.) and trifluoromethylsulfonic acid (1 eq.) in thioanisole at room temperature (yield 82%)

4) oxidation of 2-(3-mercaptophenyl) 2-methyl propionitrile in DMSO gave di-[3-(1-cyano-1-methylethyl)phenyl] disulfide (yield: 73%)

Example 16

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(2-chloro-4-fluorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1 except that O-(t-butyldimethylsilyl)hydroxylamine (1.1 equivalent) was used instead of hydroxylamine hydrochloride and 2,6-lutidine (1 equivalent) was used as a base, from N^2-[4-hydroxy-2S-isobutyl-3S-(2-chloro-4-fluorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (1 g, 2.1 mmol) there was obtained N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(2-chloro-4-fluoro-phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (451 mg, 45%) as a white solid after purification of the crude mixture by C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (gradient from 20/80 to 65/35): ^{1}H-NMR (DMSO d-6): 0.78 (d, 3H, J= 6.6 Hz), 0.86 (d, 3H, J= 6.6 Hz), 0.92 (s, 9H), 1.03 (m, 1H), 1.37 (m, 1H), 1.49 (m, 1H), 2.56 (d, 3H, J= 4.8 Hz), 3.69 (d, 1H, J= 11 Hz), 4.20 (d, 1H, J= 9.5 Hz), 7.23 (td, 1H, J_t= 8.8 Hz, J_d= 2.9 Hz), 7.46 (dd, 1H, J= 2.9 Hz, J'= 8.8 Hz), 7.80 (dd, 1H, J= 4 Hz, J'= 8.8 Hz), 9.03 (s, 1H), 10.72 (s, 1H); MS (ESI): 498 (M ^{35}Cl + Na^+), 500 (M ^{37}Cl + Na^+).

The starting material was prepared as follows:

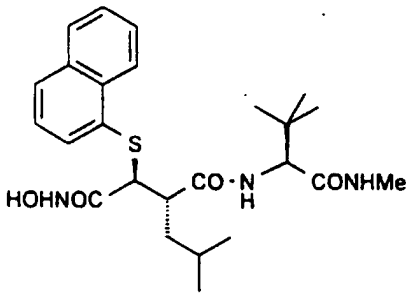
(i) In a manner analogous to that described in Example 1 (i) , from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (2.7 g, 11.8 mmol) and di-[2-chloro-4-fluorophenyl] disulfide[2] (4.2 g, 13 mmol), there was obtained 2S-isobutyl-3S-(2-chloro-4-fluorophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (2.5 g, 54 %): MS (ESI): 413 (M ^{35}Cl + Na^+), 415 (M ^{37}Cl + Na^+). Further elution gave 2S-isobutyl-3R-(2-chloro-4-fluorophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester.

(ii) In a manner analogous to that described in Example 1 (ii) except that no DMAP was added, from 2S-isobutyl-3S-(2-chloro-4-fluorophenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (2.5 g, 6.4 mmol), there was obtained N^2-[2S-isobutyl-3S-(2-chloro-4-fluorophenyl)thio 4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (2.38 g, 72%) as a white foam. MS (ESI): 539 (M ^{35}Cl + Na^+), 541 (M ^{37}Cl + Na^+).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(2-chloro-4-fluorophenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (2.3 g, 4.4 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(2-chloro-4-fluorophenyl)thio succinyl]-L-tert-leucine-N^1-methylamide (2.0 g, 97%) as a white solid: MS (ESI): 483 (M ^{35}Cl + Na^+), 485 (M ^{37}Cl + Na^+).

Example 17

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(naphth-1-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1, except that O-(t-butyldimethylsilyl)hydroxylamine (1.3 equivalents) was used instead of hydroxylamine hydrochloride and 2,6-lutidine (1.5 equivalents) was used, from N^2-[4-hydroxy-2S-isobutyl-3S-(naphth-1-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (1.0 g, 2.18 mmol) there was obtained after treatment of the crude reaction mixture with HCl (2N, 1.5 ml) and purification by C18 preparative HPLC using as eluant a mixture of acetonitrile and water/1% AcOH (gradient from 20/80 to 50/50), N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(naphth-1-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (240 mg, 24%) as a white solid: ^{1}H-NMR (DMSO d-6): 0.76 (d, 3H, J= 6.2 Hz), 0.87 (d, 3H, J= 6.2 Hz), 0.97 (s, 9H), 1.0 (m, 1H), 1.35-1.55 (m, 2H), 2.57 (d, 3H, J= 4.0 Hz), 3.15 (m, 1H), 3.61 (d, 1H, J= 11.4 Hz), 4.31 (d, 1H, J= 9.1 Hz), 7.45 (m, 1H), 7.53 (m, 2H), 7.69 (d, 1H, J= 7.3 Hz), 7.83 (q, 1H, J= 4.0 Hz), 7.87 (d, 1H, J= 8.0 Hz), 7.93 (m, 1H), 8.09 (d, 1H, J= 9.1 Hz), 8.32 (m, 1H), 8.88 (s, 1H), 10.43 (s br, 1H); MS (ESI): 496 ($M + Na^+$).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i), from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (2.0 g, 8.7 mmol) and di-(naphth-1-yl) disulfide[7] (3.0 g), there was obtained 2S-isobutyl-3S-(naphth-1-yl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.3g, 41%): MS (EI): 388 (M^+); the other isomer 2S-isobutyl-3R-(naphth-1-yl)thiobutan-1,4-dioic acid-4-tert-butyl ester which was formed in the reaction as a minor component was not isolated.

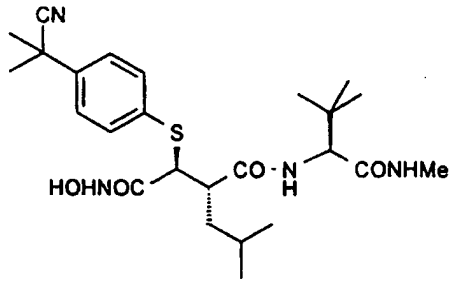
(ii) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3S-(naphth-1-yl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.3 g, 3.3 mmol), there was obtained N^2-[2S-isobutyl-3S-(naphth-1-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.26 g, 74%) as a white solid: MS (ESI): 537 ($M + Na^+$).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(naphth-1-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.2 g, 2.3 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(naphth-1-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (1.05 g, 100%): MS (ESI): 459 ($M + H^+$).

[7]Prepared by oxidation of the commercially available thiol in DMSO.

Example 18

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(4-(1-cyano-1-methylethyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1, except that O-(t-butyldimethylsilyl)hydroxylamine (1.1 equivalents) was used instead of hydroxylamine hydrochloride, from N^2-[4-hydroxy-2S-isobutyl-3S-(4-(1-cyano-1-methylethyl)phenyl)-thiosuccinyl]-L-tert-leucine-N^1-methylamide (1.4 g, 2.9 mmol) there was obtained after treatment of the crude reaction mixture with HCl (2N, 3.0 ml) and purification by C18 preparative HPLC using as eluant a mixture of methanol and water/1% AcOH (gradient from 20/80 to 60/40), N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(4-(1-cyano-1-methylethyl)phenyl)-thiosuccinyl]-L-tert-leucine-N^1-methylamide (140 mg, 10%) as a white solid: m.p. 233-236°C: ^{1}H-NMR (DMSO d-6): 0.77 (d, 3H, J= 6.2 Hz), 0.85 (d, 3H, J= 6.2 Hz), 0.95 (s, 9H), 1.0 (m, 1H), 1.3-1.5 (m, 2H), 1.68 (s, 6H), 2.56 (d, 3H, J= 4.4 Hz), 3.05 (m, 1H), 3.58 (d, 1H, J= 11.4 Hz), 4.24 (d, 1H, J= 9.1 Hz), 7.42 (m, 4H), 7.81 (q, 1H, J=4.4 Hz), 8.03 (d, 1H, J= 9.1 Hz), 9.02 (s br, 1H), 10.68 (s br, 1H); MS (ESI): 513 (M + Na^+), 529 (M + K^+). The other isomer N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(4-(1-cyano-1-methylethyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide which was formed in the reaction was not separated from the first isomer and was isolated as a mixture (710 mg, 51%) therewith.

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i), from 2R-isobutylbutan-1,4-dioic acid 4-tert-butyl ester (2.5 g, 10.8 mmol) and di-4-(1-cyano-1-methylethyl)phenyl disulfide[8]

(4.2 g), there was obtained 2S-isobutyl-3S-(4-(1-cyano-1-methylethyl)phenyl)thiobutan-1,4-dioic acid 4-tert-butyl ester (1.6g, 38%): ^{1}H-NMR ($CDCl_3$): 0.91 (dd, 6H, J= 6.6 Hz), 1.35 (m, 1H), 1.40 (s, 9H), 1.63 (m, 1H), 1.71 (s, 6H), 1.73 (m, 1H), 2.97 (m, 1H), 3.66 (d, 1H, J= 10.3 Hz), 7.46 (m, 4H); the other isomer 2S-isobutyl-3R-(4-(1-cyano-1-methylethyl)-phenyl)thiobutan-1,4-dioic acid 4-tert-butyl ester which was formed in the reaction as a minor component was not isolated.

(ii) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3S-(4-(1-cyano-1-methylethyl)phenyl)thiobutan-1,4-dioic acid-4-tert-butyl ester (1.6 g, 3.9 mmol), there was obtained N^2-[2S-isobutyl-3S-(4-(1-cyano-1-methylethyl)phenyl)thio-4-tert-butyloxy-succinyl]-L-tert-leucine-N^1-methylamide (1.6 g, 77%) as a gum: MS (ESI): 554 ($M + Na^+$).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(4-(1-cyano-1-methylethyl)phenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (1.2 g, 2.3 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(4-(1-cyano-1-methylethyl)-phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (1.4 g, 100%) as a solid: ^{1}H-NMR (DMSO d-6): 0.79 (d, 3H, J= 6.6 Hz), 0.88 (d, 3H, J= 6.6 Hz), 0.96 (s, 9H), 1.06 (m, 1H), 1.4 (m, 1H), 1.55 (m, 1H), 1.68 (s, 6H), 2.58 (d, 3H, J= 4.4 Hz), 3.06 (m, 1H), 3.64 (d, 1H, J= 11.4 Hz), 3.7 (m br 1H), 4.26 (d, 1H, J= 9.1 Hz), 7.46 (m, 4H), 7.87 (q, 1H, J= 4.4 Hz), 8.13 (d, 1H, J= 9.1 Hz).

[8]Di-4-(1-cyano-1-methylethyl)phenyl disulfide was prepared as follows:

1) alkylation of 4-bromophenylacetonitrile with excess sodium hydride/methyl iodide in THF gave 2-(4-bromophenyl)-2-methylpropionitrile (yield: 90%)

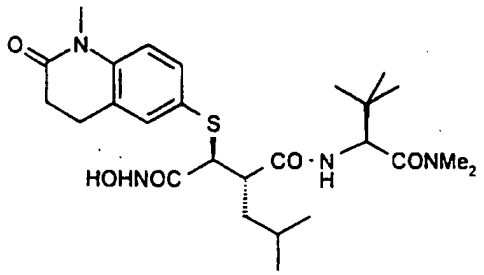
2) 2-(4-tert-butylthiophenyl)-2-methylpropionitrile was obtained from 2-(4-bromophenyl)-2-methylpropionitrile by reaction with tert-butyl mercaptan (1.1 eq.), potassium tert-butoxide (1.35 eq.) in DMSO at 70°C in the presence of $Pd(PPh_3)_4$ (0.03 eq) (yield: 40%)

3) 2-(4-mercaptophenyl)-2-methylpropionitrile was obtained from 2-(4-tert-butylthiophenyl)-2-methylpropionitrile by reaction with trifluoroacetic acid (1.7 eq.) and trifluoromethylsulfonic acid (1 eq.) in thioanisole at room temperature (yield 77%)

4) oxidation of 2-(4-mercaptophenyl)-2-methylpropionitrile in DMSO gave di-4-(1'-cyano-1'-methylethyl)phenyl disulfide (yield: 94%)

Example 19

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-dimethylamide



In a manner analogous to that described in the first paragraph of Example 1, except that O-(t-butyldimethylsilyl)hydroxylamine (1.1 equivalents) was used instead of hydroxylamine hydrochloride, from N^2- [4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-dimethylamide (450 mg, 0.89 mmol) there was obtained after treatment of the crude reaction mixture with HCl (6N, 0.2 ml) and purification by C18 preparative HPLC using as eluant a mixture of methanol and water/1% AcOH (gradient from 20/80 to 60/40), N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-dimethylamide (274 mg, 59%) as a white solid: m.p. 240-242°C: ^{1}H-NMR (DMSO d-6): 0.76 (d, 3H, J= 6.6 Hz), 0.83 (d, 3H, J= 6.6 Hz), 1.0 (s, 9H), 1.0 (m, 1H), 1.3-1.5 (m, 2H), 2.53 (m, 2H), 2.82 (s, 3H), 2.83 (m, 2H), 3.05 (m, 1H), 3.09 (s, 3H), 3.24 (s, 3H), 3.45 (d, 1H, J= 11.4 Hz), 4.81 (d, 1H, J= 9.2 Hz), 7.03 (d, 1H, J= 8.8 Hz), 7.22 (s, 1H), 7.27 (dd, 1H, J=1.8 Hz, J=8.8 Hz), 8.09 (d, 1H, J= 9.2 Hz), 8.96 (s br, 1H), 10.61 (s br, 1H); MS (ESI): 543 (M + Na^+).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiobutan-1,4-dioic acid 4-tert-butyl ester (400 mg, 0.95

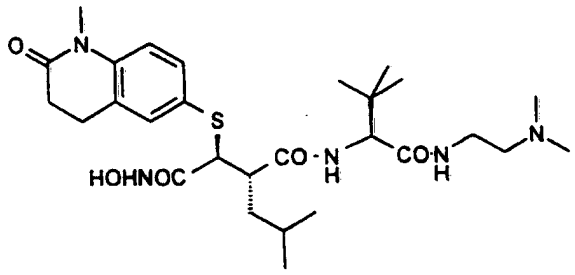
mmol), described in Example 9, and L-tert-leucine dimethylamide[9] (165mg, 1.04 mmol) there was obtained N^2-[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-dimethylamide (500 mg, 94%) as a white solid: MS (ESI): 584 ($M + Na^+$).

(ii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-dimethylamide (500 mg, 0.89 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-dimethylamide (450 mg, 100%) as a solid: ^{1}H-NMR (DMSO d-6): 0.78 (d, 3H, J= 6.6 Hz), 0.86 (d, 3H, J= 6.6 Hz), 1.0 (s, 9H), 1.25 (m, 1H), 1.3-1.6 (m, 2H), 2.53 (m, 2H), 2.82 (s, 3H), 2.83 (m, 2H), 3.05 (m, 1H), 3.1 (s, 3H), 3.24 (s, 3H), 3.53 (d, 1H, J= 11.4 Hz), 4.6 (s br, 1H), 4.86 (d, 1H, J= 9.1 Hz), 7.06 (d, 1H, J= 8.8 Hz), 7.26 (s, 1H), 7.33 (m, 1H), 8.19 (d, 1H, J= 9.1 Hz).

[9] L-tert-leucine dimethylamide was prepared by the reaction of L-tert-leucine with triphosgene to give 3-(S)-tert-butyl oxazolidine-1,4-dione which was then treated with a saturated solution of dimethylamine in ether.

Example 20

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-(2-dimethylamino)ethylamide



In a manner analogous to that described in the first paragraph of Example 1, except that O-(t-butyldimethylsilyl)hydroxylamine (1.1 equivalents) was used instead of hydroxylamine hydrochloride, from N^2- [4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydro-quinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-(2-dimethylamino)ethylamide (310 mg, 0.56 mmol) there was obtained after treatment of the crude reaction mixture with HCl (2N, 0.5 ml) and purification by C18 preparative HPLC using as eluant a mixture of methanol and water/1% AcOH (gradient from 15/85 to 50/50), N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-(2-dimethyl-amino)ethylamide (123 mg, 39%) as a white solid: m.p. 152-158°C; ^{1}H-NMR (DMSO d-6): 0.77 (d, 3H, J= 6.6 Hz), 0.86 (d, 3H, J= 6.6 Hz), 0.99 (s, 9H), 1.1 (m, 1H), 1.3-1.5 (m, 2H), 2.5-2.57 (m, 8H), 2.7-2.75 (m, 2H), 2.83 (m, 2H), 3.04 (m, 1H), 3.24 (s, 3H), 3.33-3.4 (m, 2H), 3.47 (d, 1H, J= 11.4 Hz), 4.24 (d, 1H, J= 8.8 Hz), 7.02 (d, 1H, J= 8.4 Hz), 7.22 (s, 1H), 7.26 (dd, 1H, J= 1.8 Hz, J= 8.4 Hz), 8.1 (m, 2H), 8.98 (s br, 1H), 10.57 (s br, 1H); MS (ESI): 564 ($M + H^+$).

The starting material was prepared as follows:

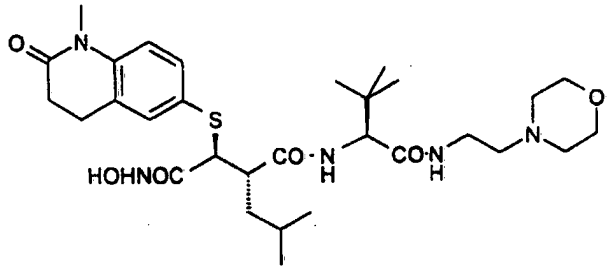
(i) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiobutan-1,4-dioic acid 4-tert-butyl ester (400 mg, 0.95 mmol), described in Example 9, and L-tert-leucine (2-dimethylamino)ethylamide[10] (210mg, 1.04 mmol) there was obtained N^2-[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydro-quinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-(2-dimethylamino)ethylamide (369 mg, 64%) as a white solid: MS (ESI): 605 ($M + H^+$).

(ii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-(2-dimethylamino)ethylamide (324 mg, 0.56 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-(2-dimethylamino)ethylamide (430 mg, 100%) as a cream solid: MS (ESI): 548 (M^+).

[10] L-tert-leucine 2-(dimethylamino)ethylamide was prepared by the reaction of L-tert-leucine with triphosgene to give 3-(S)-tert-butyloxazolidine-1,4-dione which was then treated with N,N-dimethyl ethylenediamine.

Example 21

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-[2-(4-morpholino)ethyl]amide



In a manner analogous to that described in the first paragraph of Example 1, except that O-(t-butyldimethylsilyl)hydroxylamine (1.1 equivalents) was used instead of hydroxylamine hydrochloride, from N^2-[4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydro-quinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-[2-(4-morpholino)ethyl]amide (237 mg, 0.4 mmol) there was obtained after treatment of the crude reaction mixture with HCl (2N, 0.5 ml) and purification by C18 preparative HPLC using as eluant a mixture of methanol and water/1% AcOH (gradient from 15/85 to 50/50), N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-[2-(4-morpholino)ethyl]amide (85 mg, 35%) as a white solid: m.p. 156-158°C ; ^{1}H-NMR (DMSO d-6 + TFA) : 0.78 (d, 3H, J = 6.3 Hz), 0.87 (d, 3H, J = 6.3 Hz), 1.0 (s, 9H), 1.0 (m, 1H), 1.41 (m, 2H), 2.51 (m, 2H), 2.84 (m, 2H), 3.08-3.24 (m, 5H), 3.24 (s, 3H), 3.4-3.5 (m, 4H), 3.45 (d, 1H, J = 11.3 Hz), 3.65 (m, 2H), 4.0 (m, 2H), 4.12 (m, 1H), 7.01 (d, 1H, J = 8.5 Hz), 7.21 (d, 1H, J = 2.2 Hz), 7.26 (dd, 1H, J = 2.2 Hz, J = 8.5 Hz), 8.18 (d, 1H, J = 7.8 Hz); MS (ESI) : 606 ($M + H^+$).

The starting material was prepared as follows:

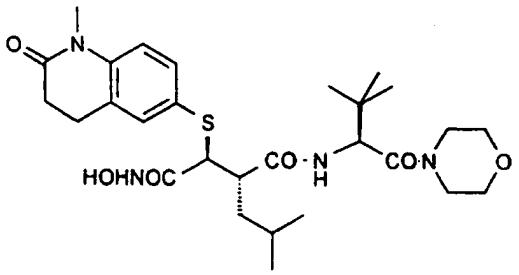
(i) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiobutan-1,4-dioic acid 4-tert-butyl ester (300 mg, 0.71 mmol), described in Example 9, and L-tert-leucine 2-(4-morpholino)ethylamide[(11)] (190 mg, 0.78 mmol) there was obtained N^2-[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-[2-(4-morpholino)ethyl]amide (270 mg, 58%) as a white solid: MS (ESI) : 647 ($M + H^+$) and 669($M + Na^+$).

(ii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-[2-(4-morpholino)ethyl]amide (260 mg, 0.4 mmol) there was obtained N^2-[4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-[2-(4-morpholino)ethyl]amide (324 mg, 100%) : MS (ESI) : 591 ($M + H^+$).

[(11)]L-tert-leucine 2-(4-morpholino)ethylamide was prepared by the reaction of L-tert-leucine with triphosgene to give 3-(S)-tert-butyloxazolidine-1,4-dione which was then treated with 4-(2-aminoethyl)morpholine.

Example 22

N^2-[[4-(N-Hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine]-N^1-(4-morpholine)amide



In a manner analogous to that described in the first paragraph of Example 1, except that O-(t-butyldimethylsilyl)hydroxylamine (1.1 equivalents) was used instead of hydroxylamine hydrochloride, from N^2-[[4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-

tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine]-N^1-(4-morpholine)amide (415 mg, 0.76 mmol) there was obtained after treatment of the crude reaction mixture with HCl (2N, 0.5 ml) and purification by C18 preparative HPLC using as eluant a mixture of methanol and water/1% AcOH (gradient from 15/85 to 50/50), N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine]-N^1-(4-morpholine)amide (137 mg, 32%) as a white powder : m.p. 218-220°C ; ^{1}H-NMR (DMSO d-6) : 0.76 (d, 3H, J = 6.6 Hz), 0.82 (d, 3H, J = 6.6 Hz), 0.94 (m, 1H), 0.99 (s, 9H), 1.35-1.5 (m, 2H), 2.51-2.55 (m, H), 2.8-2.85 (m, 2H), 3.02 (m, 1H), 3.22 (s, 3H), 3.32-3.4 (m, 2H), 3.45 (d, 1H, J = 11.3 Hz), 3.55-3.75 (m, 6H), 4.82 (d, 1H, J = 8.8 Hz), 7.0 (d, 1H, J = 8.4 Hz), 7.2 (d, 1H, J = 1.8 Hz), 7.25 (dd, 1H, J = 1.8 Hz, J = 8.4 Hz), 8.16 (d, 1H, J = 9.1 Hz), 8.95 (s br, 1H), 10.57 (s br, 1H) ; MS (ESI) : 563 (M + H$^+$) and 585 (M + Na$^+$).

The starting material was prepared as follows:

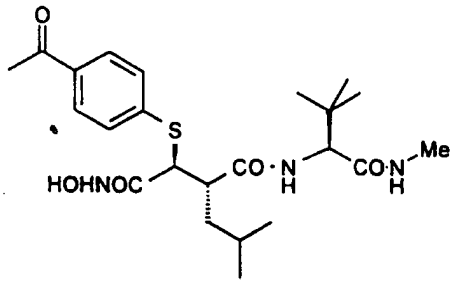
(i) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thio-butan-1,4-dioic acid 4-tert-butyl ester (375 mg, 0.89 mmol), described in Example 9, and N-(L-tert-leucine)-(4-morpholine)amide [12] (196 mg, 0.98 mmol) there was obtained N-[[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydro-quinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine]-N^1-(4- morpholine)amide (465 mg, 86%) as a white solid: MS (ESI) : 626 (M + Na$^+$).

(ii) In a manner analogous to that described in Example 1 (iii), from N-[[2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine]-N^1-(4-morpholine)amide (460 mg, 0.76 mmol) there was obtained N-[[4-hydroxy-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine]-N^1-(4-morpholine (417 mg, 100%) : MS (ESI) : 548 (M + H$^+$), 570 (M + Na$^+$).

[12] N-(L-tert-leucine)-(4-morpholine)amide was prepared by the reaction of L-tert-leucine with triphosgene to give 3-(S)-tert-butyloxazolidine-1,4-dione which was then treated with morpholine.

Example 23

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(4-acetylphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide



In a manner analogous to that described in the first paragraph of Example 1, except that O-(t-butyldimethylsilyl) hydroxylamine (1.2 equivalents) was used instead of hydroxylamine hydrochloride, from a mixture of N^2- [4-hydroxy-2S-isobutyl-3S-(4-acetylphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide and the corresponding 3R epimer (400 mg, 0.89 mmol), there was obtained after treatment of the crude reaction mixture with HCl (2N, 0.5 ml) and purification by C18 preparative HPLC using as eluant a mixture of methanol and water/1% AcOH (gradient from 30/70 to 60/40), N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(4-acetylphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (80 mg, 20%) as a white solid : m.p. 210-211°C; ^{1}H-NMR (DMSO d-6) : 0.79 (d, 3H, J = 6.6 Hz), 0.87 (d, 3H, J = 6.6 Hz), 0.9 (s, 9H), 1.04 (m, 1H), 1.39 (m, 1H), 1.49 (m, 1H), 2.56 (s, 3H), 2.57 (d, 3H, J= 3.7 Hz), 3.09 (m, 1H), 3.82 (d, 1H, J = 11.0 Hz), 4.2 (d, 1H, J = 9.2 Hz), 7.46 (d, 2H, J = 8.4 Hz), 7.8 (m, 1H), 7.85 (d, 2H, J = 8.4 Hz), 8.07 (d, 1H, J = 9.2 Hz), 9.07 (s br, 1H), 10.89 (s br, 1H) ; MS (ESI) : 488 ($M + Na^+$),

and the other epimer, N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(4-acetylphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide (183 mg, 44%) as a white solid : m.p. 220-221°C ;
MS (ESI) : 466 ($M + H^+$), 488 ($M + Na^+$).

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i), from 2R-isobutylbutan-1,4-dioic acid 4-tert-butyl ester (5.0 g, 21.7 mmol) and di-[4-(2-methyl-1,3-dioxolan-2-yl)phenyl] disulfide[13] (9.32 g), there was obtained a mixture of 2S-isobutyl-3S-[4-(2-methyl-1,3-

dioxolan-2-yl)phenyl]thiobutan-1,4-dioic acid 4-tert-butyl ester and the corresponding 3R epimer (5.53g, 60%) (3S:3R, 1:3) : MS (ESI): 447 ($M + Na^+$).
This mixture was used unseparated for the next steps.

(ii) In a manner analogous to that described in Example 1 (ii), from the mixture of 2S-isobutyl-3S-[4-(2-methyl-1,3-dioxolan-2-yl)phenyl]thiobutan-1,4-dioic acid 4-tert-butyl ester and the corresponding 3R epimer (2.33 g, 5.49 mmol), there was obtained a mixture of N^2-[2S-isobutyl-3S-{4-(2-methyl-1,3-dioxolan-2-yl)phenyl}thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide together with the corresponding 3R isomer (2.4 g, 79%) as an off white solid: MS (ESI): 551 ($M + H^+$). 573 ($M + Na^+$), 589 ($M + K^+$).

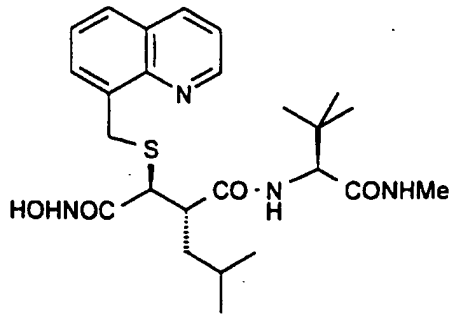
(iii) Hydrochloric acid (2N, 800 µl) was added to a solution of the mixture of N^2-[2S-isobutyl-3S-{4-(2-methyl-1,3-dioxolan-2-yl)phenyl}thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide together with the corresponding 3R isomer (1.74 g, 3.16 mmol) in acetone (17 ml) and the mixture was stirred at room temperature for 4 hours. The acetone was evaporated, the residue taken up in dichloromethane, washed with water and brine and dried over $MgSO_4$. The solvent was removed in vacuo to give a mixture of N^2-[2S-isobutyl-3S-(4-acetylphenyl)-thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide together with the corresponding 3R isomer (1.52 g, 79%) as an off white solid: MS (ESI): 507 ($M + H^+$), 529 ($M + Na^+$).

(iv) In a manner analogous to that described in Example 1 (iii), from mixture of N^2-[2S-isobutyl-3S-(4-acetylphenyl)thio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide together with the corresponding 3R isomer (1.5 g, 2.96 mmol) there was obtained a mixture of N^2-[4-hydroxy-2S-isobutyl-3S-(4-acetylphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide and the corresponding 3R epimer (1.17 g, 88%) :MS (ESI): 451 ($M + H^+$), 473 ($M + Na^+$).

(13) Zeneca Ltd., (Bird, T. G. C.; Ple, P.), Patent EP0555068 (1993), in Example 7.

Example 24

N^2-[4-(N-Hydroxyamino)-2S-isobutyl-3S-(quinolin-8-yl)methylthiosuccinyl]-L-tert-leucine-N^1-methylamide



To a solution of N^2-[4-hydroxy-2S-isobutyl-3(R/S)-(quinolin-8-yl)methylthiosuccinyl]-L-tert-leucine-N^1-methylamide (460 mg) in DMF (5 ml) cooled at 0°C were added 1-hydroxybenzotriazole (270 mg), N-methylmorpholine (400 µl), O-(2,4-dimethoxybenzyl)hydroxylamine[12] (366 mg) and N-ethyl-N'-(3-dimethylaminopropyl)carbodiimide hydrochloride (382 mg). The mixture was stirred at room temperature for 18 hours. The mixture was partitioned between water and ethyl acetate. The organic layer was washed with saturated sodium bicarbonate, brine, dried over $MgSO_4$ and filtered. The solvents were evaporated in vacuo to give crude N^2-[4-(N-(2,4-dimethoxybenzyloxy)amino)-2S-isobutyl-3(R/S)-(quinolin-8-yl)methylthiosuccinyl]-L-tert-leucine-N^1-methylamide. This crude material was dissolved in dichloromethane (10 ml). Trifluoroacetic acid (0.7 ml) was added. The mixture was stirred at room temperature for 15 minutes. The solvents were evaporated in vacuo. Methanol was added (20 ml) and the solids were filtered off. The filtrate was concentrated and purified by C18 preparative HPLC using as eluant a mixture of methanol and water/1%AcOH (gradient from 0/100 to 50/50) to give the title compound (76 mg): ^{1}H-NMR (DMSO d-6): 0.78 (d, 3H, J= 6.6 Hz), 0.86 (m, 12H), 1.04 (m, 1H), 1.5-1.3 (m, 2H), 2.56 (d, 3H, J= 4.4 Hz), 3.08 (m, 1H), 3.28 (d, 1H, J= 11 Hz), 4.20 (d, 1H, J= 9.2 Hz), 4.43 (d, 1H, J= 12.5 Hz), 4.54 (d, 1H, J= 12.5 Hz), 7.55 (m, 2H), 7.80 (m, 2H), 7.89 (m, 2H), 8.36 (m, 1H), 8.93 (m, 1H), 9.04 (s, 1H), 10.75 (s, 1H); MS (EI): 488 (M^+).

[12] Prepared from 2,4-dimethoxybenzyl alcohol according to Grochowski E, Jurczak J; Synthesis, 1976, 682.

The starting material was prepared as follows:

(i) In a manner analogous to that described in Example 1 (i), from 2R-isobutylbutan-1,4-dioic acid-4-tert-butyl ester (400 mg, 1.74 mmol) and di-(quinolin-8-yl)methyl disulfide[13] (726 mg, 2.08 mmol), there was obtained 2S-isobutyl-3(R/S)-(quinolin-8-yl)methylthiobutan-1,4-dioic acid-4-tert-butyl ester (487 g, 1:1) contaminated with ca 20% of starting carboxylic acid.
MS (ESI): 404 ($M + H^+$).

[13a] Prepared from 8-mercaptomethylquinoline (Chem. Abstracts, 1961, 14458h)

(ii) In a manner analogous to that described in Example 1 (ii), from 2S-isobutyl-3(R/S)-(quinolin-8-yl)methylthiobutan-1,4-dioic acid-4-tert-butyl ester (450 mg, 1.12 mmol), there was obtained N^2-[2S-isobutyl-3(R/S)-(quinolin-8-yl)methylthio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (451 mg, 1:1).

(iii) In a manner analogous to that described in Example 1 (iii), from N^2-[2S-isobutyl-3(R/S)-(quinolin-8-yl)methylthio-4-tert-butyloxysuccinyl]-L-tert-leucine-N^1-methylamide (450 mg) there was obtained N^2-[4-hydroxy-2S-isobutyl-3(R/S)-(quinolin-8-yl)methylthiosuccinyl]-L-tert-leucine-N^1-methylamide as the trifluoroacetate salt : MS (EI): 474 ($M + H^+$).

Example 25

Typical tablet formulations for a compound of this invention or a pharmaceutically-acceptable salt thereof ('Compound X') are:

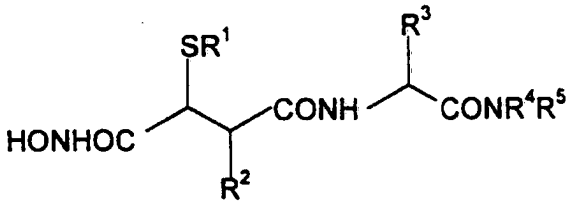
(a)	Tablet Formulation 1	mg/tablet
	Compound X.....	100
	Lactose Ph.Eur.....	179
	Croscarmellose sodium.....	12
	Polyvinylpyrrolidone.....	6
	Magnesium stearate.....	3

(b)	Tablet Formulation II	mg/tablet
	Compound X	250
	Lactose Ph.Eur	215
	Croscarmellose sodium	20
	Polyvinylpyrrolidone	10
	Magnesium stearate	5

The tablets may be prepared by conventional procedures well known in the pharmaceutical art and may be film coated with typical coating materials such as hydroxypropylmethylcelluose.

CLAIMS

A compound of the formula (I):



(I)

wherein:

R^1 is aryl, arylC_{1-6}alkyl, heteroaryl or heteroarylC_{1-6}alkyl;

R^2 is hydrogen, C_{1-8}alkyl, C_{2-6}alkenyl, C_{2-6}alkynyl, C_{3-8}cycloalkyl, heteroaryl, heterocyclyl, arylC_{1-6}alkyl, heteroarylC_{1-6}alkyl, heterocyclylC_{1-6}alkyl or C_{3-8}cycloalkylC_{1-6}alkyl;

R^3 is C_{1-6}alkyl, C_{2-6}alkenyl, aryl, C_{1-6}alkyl, heteroarylC_{1-6}alkyl or the side-chain of a naturally occurring amino acid;

R^4 is hydrogen, C_{1-6}alkyl, C_{3-8}cycloalkyl, C_{4-8}cycloalkenyl, arylC_{1-6}alkyl, heteroarylC_{1-6}alkyl or heterocyclylC_{1-6}alkyl;

R^5 is hydrogen or C_{1-6}alkyl; or R^4 and R^5 together with the nitrogen atom to which they are joined form a heterocyclic ring;

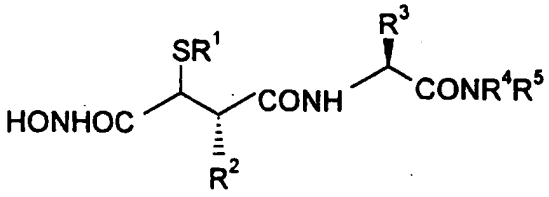
wherein any group or ring, in R^1-R^5, is optionally substituted;

or a pharmaceutically acceptable salt or an in vivo hydrolysable ester thereof.

2. A compound according to claim 1 wherein R^1 is phenyl, phenylC_{1-6}alkyl, naphthylC_{1-6}alkyl, heteroaryl or heteroarylC_{1-6}alkyl wherein any of such rings is unsubstituted or substituted by one or two groups selected from halogen, C_{1-6}alkylcarbonyl, C_{1-6}alkylsulfonyl, trifluoromethyl, cyano, C_{1-6}alkyl, C_{1-6}alkoxy, cyanoC_{1-6}alkyl, or two adjacent carbon atoms on a phenyl ring are linked to form a methylenedioxy group.

3. A compound according to claim 1 wherein R^1 is phenyl and two adjacent carbon atoms are linked by $-(CH_2)_m-$ wherein m is 3 or 4, by $-NR^a-CO-(CH_2)_n-$ wherein R is hydrogen or C_{1-6}alkyl and n is 1 or 2, by $-NR^a-COCH=CH-$, $-CO-NR^a-(CH_2)_n-$ or by $-CONR^a-CH=CH-$.

4. A compound according to claim 1 which is of the formula (II):



(II)

wherein R^1 is phenyl unsubstituted or substituted by one or two groups selected from halogen C_{1-6}alkylsulphonyl, trifluoromethyl, C_{1-6}alkyl, C_{1-6}alkoxy, cyano, C_{1-6}alkanoyl, cyanoC_{1-6}alkyl, or two adjacent carbon atoms on the phenyl ring are linked to form a methylenedioxy ($-OCH_2O-$) group; R^2 is isobutyl; R^3 is isobutyl, tert-butyl, 1,1-dimethylmethylthiomethyl or benzyl; R^4 is methyl, ethyl, n-propyl, isobutyl, tert-butyldimethylaminoethyl or benzyl; and R^5 is hydrogen or methyl, or R^4 and R^5 together with the nitrogen atom to which they are joined form a morpholino ring.

5. A compound according to claim 1 wherein R^1 is quinolinyl, isoquinolinyl, 1-methyl-2-oxodihydroquinolinyl, 1-methyl-2-oxotetrahydroquinolinyl, 2-methyl-1-oxodihydro-isoquinolinyl or 2-methyl-1-oxotetrahydroisoquinolinyl; R^2 is isobutyl; R^3 is isobutyl, tert-butyl, 1,1-dimethylmethylthiomethyl or benzyl; R^4 is methyl, ethyl, n-propyl, isobutyl, tert-butyl, dimethylaminoethyl or benzyl; and R^5 is hydrogen or methyl, or R^4 and R^5 together with the nitrogen atom to which they are joined form a morpholine ring.

6. A compound according to claim 1 which is:

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-phenylthiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(4-fluorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(3,5-dichlorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(3,4-dimethoxyphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(3,4-dimethoxyphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(4-(methylsulfonyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(4-(methylsulfonyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(4-cyanophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(4-cyanophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(3,5-di-(trifluoromethyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-benzylthiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(2-benzothiazol-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(6-hydroxynaphth-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3R-(6-hydroxynaphth-2-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(quinolin-2'-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-[3-(1-cyano-1-methylethyl)phenyl]thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(2-chloro-4-fluorophenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(naphth-1-yl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(4-(1-cyano-1-methylethyl)phenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-dimethylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-(2-dimethylaminoethyl)amide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine-N^1-[2-(4-morpholino)ethyl]amide;

N^2-[[4-(N-hydroxyamino)-2S-isobutyl-3S-(1-methyl-2-oxo-1,2,3,4-tetrahydroquinolin-6-yl)thiosuccinyl]-L-tert-leucine]-N^1-(4-morpholine)amide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(4-acetylphenyl)thiosuccinyl]-L-tert-leucine-N^1-methylamide;

N^2-[4-(N-hydroxyamino)-2S-isobutyl-3S-(quinolin-8-yl)methylthiosuccinyl]-L-tert-leucine-N^1-methylamide;

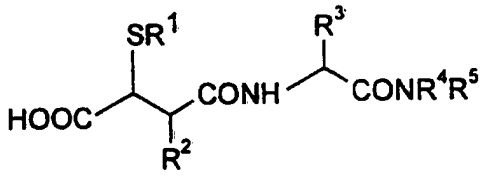
or a pharmaceutically acceptable salt thereof.

7. A pharmaceutical composition which comprises a compound according to any one of claims 1 to 6 and a pharmaceutically acceptable carrier.

8. The use of a compound according to any one of claims 1 to 6 for the manufacture of a medicament for treating disease conditions mediated by TNF.

9. A process for preparing a compound according to any one of claims 1-6 or a pharmaceutically acceptable salt or in vivo hydrolysable ester thereof which process comprises

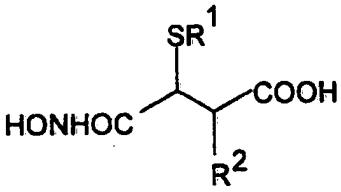
a) reacting a compound of the formula (III):



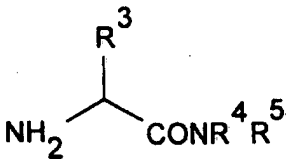
(III)

wherein R^1-R^5 are as defined in claim 1, or an activated derivative thereof with hydroxylamine, O-protected hydroxylamine or a salt thereof; or

b) coupling a compound of the formula (IV) with a compound of the formula (V):



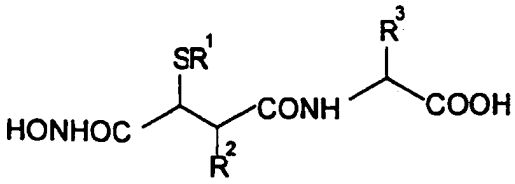
(IV)



(V)

wherein R^1-R^5 are as defined in claim 1, under standard peptide coupling conditions; or

c) reacting a compound of the formula (VI) with compound of the formula (VII):



(VI)

HNR^4R^5 (VII)

wherein R^1-R^5 are as defined in claim 1,

wherein any functional group is protected, if necessary, and:

i. removing any protecting groups;

ii. optionally forming a pharmaceutically acceptable salt or in vivo hydrolysable ester.

10. A compound of the formula (III) as defined in claim 9.

INTERNATIONAL SEARCH REPORT

International Application No
PCT/GB 97/01164

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 C07C323/22 C07C323/60 C07C323/62 A61K31/16 A61K31/255

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 C07K C07C A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 90 05719 A (BIRITISH BIOTHECH PHARMACEUTICALS LIM., OXFORD, GB) 31 May 1990	1,2
Y	whole document, especially examples and claims	3-10

X	WO 95 09841 A (BIRITISH BIOTHECH PHARMACEUTICALS LIM., OXFORD, GB) 13 April 1995	1,2
Y	whole document, especially examples and claims	3-10

X	WO 94 10990 A (BIRITISH BIOTHECH PHARMACEUTICALS LIM., OXFORD, GB) 26 May 1994	1-10
	whole document, especially claim 13	

	-/--	

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
14 August 1997	10.09.97
Name and mailing address of the ISA	**Authorized officer**
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+ 31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+ 31-70) 340-3016	Kronester-Frei, A

Form PCT/ISA/210 (second sheet) (July 1992)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	EP 0 613 883 A (YAMANOUCHI PHARMACEUTICAL CO. LTD., TOKYO, JP) 7 September 1994 claims, pages 9/10 ---	1-10
Y	EP 0 236 872 A (F. HOFFMAN-LA ROCHE & CO. AG, BASLE, CH) 16 September 1987 claim 15, page 8, line 29 to page 10, line 14 -----	1-10

2 .

INTER[redacted]IONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/GB 97/01164

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9005719 A	31-05-90	AU 644064 B	02-12-93
		AU 4800390 A	12-06-90
		CA 2003718 A	23-05-90
		DE 68914687 D	19-05-94
		DE 68914687 T	08-09-94
		EP 0446267 A	18-09-91
		ES 2055409 T	16-08-94
		HU 9500245 A	28-09-95
		JP 2565599 B	18-12-96
		JP 4502008 T	09-04-92
		NO 177701 B	31-07-95
		US 5310763 A	10-05-94
		US 5240958 A	31-08-93
WO 9509841 A	13-04-95	AU 7787594 A	01-05-95
		EP 0722438 A	24-07-96
		JP 9503222 T	31-03-97
		ZA 9407799 A	10-07-95
WO 9410990 A	26-05-94	AT 150300 T	15-04-97
		AU 5430194 A	08-06-94
		DE 69309094 D	24-04-97
		DE 69309094 T	31-07-97
		EP 0667770 A	23-08-95
		ES 2101358 T	01-07-97
		JP 8505605 T	18-06-96
EP 613883 A	07-09-94	JP 5125029 A	21-05-93
		US 5442110 A	15-08-95
		AU 2799792 A	07-06-93
		CA 2122046 A	13-05-93
		CN 1073166 A	16-06-93
		WO 9309090 A	13-05-93
		US 5473100 A	05-12-95
EP 236872 A	16-09-87	AU 588437 B	14-09-89
		AU 6990287 A	17-09-87
		CA 1314655 A	16-03-93
		DE 3782751 A	07-01-93
		IE 60128 B	01-06-94

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/GB 97/01164

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
EP 236872 A		JP 6029228 B	20-04-94
		JP 62230757 A	09-10-87
		US 4996358 A	26-02-91